Library
JUN 1 3 1944

Volume 34 *Number 5*

133

Phytopathology

AN INTERNATIONAL JOURNAL

May 1944

Official Organ of

The American Phytopathological Society

$6.25 per year, Canada $6.00 per year, United States and dependencies $6.50 per year, elsewhere

Office of Publication

Corner North Queen Street and McGovern Avenue, Lancaster, Pennsylvania

Entered at the postoffice in Lancaster, Pennsylvania, as second-class matter

TABLE OF CONTENTS

Bacterial wilt of tomato caused by Phytomonas solanacearum. EDWARD K. VAUGHAN 443

Some factors influencing the solubility of cuprous oxide in relation to its toxicity as a fungicide ..... E. A. MARTEN AND J. G. LEACH 459

Variation in symptoms produced by isolates of Phytomonas medicaginis var. phaseolicola ..... J. H. JENSEN AND J. E. LIVINGSTON 471

Seedling invasion of the covered smut of oats ..... PAUL F. BRANDWEIN 481

Pathogenesis of Aphanomyces cochlioides on taproots of the sugar beet ..... W. F. BUCHHOLTZ AND C. H. MEREDITH 485

The sequence of infection of a seedling stand of sugar beets by Pythium debaryanum and Aphanomyces cochlioides. W. F. BUCHHOLTZ 490

Ornithogalum mosaic ..... FLOYD F. SMITH AND PHILIP BRIERLEY 497

Phytopathological Notes 504

- Reaction of Lycopersicon spp. to spotted wilt ..... PAUL G. SMITH
- Witches' broom of beans ..... WILLIAM G. HOYMAN
- The perennial tree onion a carrier of onion-yellow-dwarf virus. PHILIP BRIERLEY AND FLOYD F. SMITH
- A simplified method of growing plants with roots in nutrient vapors ..... L. J. KLOTZ
- A note on the effects of splash injury in guayule seedlings. FREDRICK T. ADDICOTT
- A severe necrosis caused by bean-mosaic virus 4 on beans. W. J. ZAUMEYER AND L. L. HARTER

Book Review 513

Report of the 35th Annual Meeting and War Conference of the American Phytopathological Society 515

PHYTOPATHOLOGY

An International Journal
Official Organ of The American Phytopathological Society

EDITORS

HELEN HART, Editor-in-Chief, University Farm, St. Paul Minnesota.
W. H. BURKHOLDER, Cornell University, Ithaca, New York.
A. J. RIKER, University of Wisconsin, Madison, Wisconsin.
W. J. ZAUMEYER, Horticultural Station, Beltsville, Maryland.

ASSOCIATE EDITORS

A. A. DUNLAP, Agricultural Experiment Station, College Station, Texas.
J. M. HAMILTON, N. Y. Agricultural Experiment Station, Geneva, New York.
W. G. HOYMAN, University of Arizona, Tucson, Arizona.
C. O. JOHNSON, Kansas Agricultural Experiment Station, Manhattan, Kansas.
G. C. KENT, Iowa State College, Ames, Iowa.
G. L. PELTIER, University of Nebraska, Lincoln, Nebraska.
A. G. PLAKIDAS, Louisiana State University, Baton Rouge, Louisiana.
H. N. RACICOT, Experimental Farm, Ottawa, Ontario, Canada.
R. E. SMITH, University of California, Berkeley, California.
E. S. SCHULTZ, Plant Industry Station, Beltsville, Maryland.
W. W. WAGENER, 446 Phelan Building, San Francisco, California.

BUSINESS MANAGEMENT

R. M. CALDWELL, Business Manager, A.E.S. Bldg., Purdue University, Lafayette, Indiana.
S. L. HOPPERSTEAD, Advertising Manager, University of Delaware, Newark, Delaware.

NOTICES

All communications regarding subscriptions and back numbers should be sent to R. M. CALDWELL, Business Manager, A.E.S. Bldg., Purdue University, Lafayette, Indiana.

Subscription price. $6.00 per year United States and dependencies; Canada, $6.25; other countries $6.50; single numbers of the current volume, 60 cents each. The journal has been issued monthly beginning with Volume VIII, January, 1918. Prior to that it was issued bimonthly.

Back Volumes VIII to XXXII, inclusive (except Vol. VIII, Nos. 1 and 8, Vol. IX, No. 1, and Vol. X, No. 1, which are not available) unbound at $8.00 per volume. All earlier volumes obtainable only in broken numbers.

Separate Numbers will be supplied only from broken volumes, when the supply of complete volumes becomes low. Separate numbers from Volumes VIII to XXXII, $1.00 per copy; from Volumes I to VII, $2.00 per copy. The following numbers of Volumes I to VII, inclusive, are obtainable: Volume I, Nos. 1, 4, 6, and Table of Contents; Vol. II, Nos. 1, 5, and Table of Contents; Vol. III, Nos. 4, 5; Vol. VI, index only.

Thirty-Year Index (to Vols. 1–30, 1911–1940) bound. $5.50, United States. $6.00, elsewhere.

Requests to supply copies of the journal must be made within 60 days from date of issue. Date of issue January 15 and monthly thereafter.

Advertisements. Rates may be secured from the Advertising Manager. No indorsement of any statement of claims made in advertisements is assumed by this journal or by The American Phytopathological Society.

Lost Numbers. It has been customary to supply free of charge a copy of any number of PHYTOPATHOLOGY to members and subscribers in case of nondelivery for any cause. In view of present conditions throughout the world, it is not possible to guarantee delivery. The edition of the journal is limited, of course, by our funds for publication; in case of serious losses of mail addressed to foreign countries, as for instance, on ships at sea, it would be impossible to replace the lost copies. We shall make every effort to see that delivery of issues is made to all members and subscribers, but cannot assume responsibility to replace lost copies under present world conditions. This does not apply at present to domestic subscribers and members of The American Phytopathological Society.

Separates. No gratis copies supplied. A printed schedule of prices is submitted with the proof, and authors may secure separates by placing an order on the blanks provided when proof is returned.

Manuscript may be sent to the nearest member of the editorial board. Clearness, brevity, and conciseness are essential. In form and style, such as punctuation, spelling and use of italic, manuscript should conform to the best usage in recent numbers of this journal. Manuscripts should be typed on one side of the paper, double spaced, and sent unfolded. All papers longer than four printed pages should include a summary. The name and address of the institution at which the work was done should be included at the end of the summary. All footnotes should be numbered consecutively with arabic numerals except in tables, where small letters should be used. Each table should have a heading stating clearly and concisely its contents. References to literature should be made as footnotes only when five or fewer citations are given. The simplified method of citing literature used in recent numbers of this journal should be followed. References in "Literature Cited" should be listed alphabetically and numbered consecutively.

Illustrations necessarily must be limited in number. Photographs, to reproduce satisfactorily, must be of best quality. Authors who desire unusual numbers of illustrations or special types of illustrations will be asked to bear part of the cost, i.e. in excess of $12.50, the approximate cost of 2 full-page cuts. All illustrations are reproduced on the same grade of paper and will be run as text figures unless authors specify otherwise. Since plates are printed on the same kind and quality of paper, they offer no advantage over full-page figures. There is, therefore, nothing to be gained through publication of illustrations as plates rather than as figures. Individual illustrations within a figure—or plate—should be designated by letters to avoid confusion with text figures, designated by arabic numerals. Full-page figures—or plates—should be arranged to conform to journal page size and shape ($4\frac{1}{2}'' \times 7''$) after making due allowance for reductions. Specific reference to each illustration must be made in the text. Each illustration must be accompanied by an adequate legend, the legend to be typed on separate paper and not on the illustration.

The responsibility for statements, whether of fact or opinion, rests entirely with the writers thereof.

The American Phytopathological Society

FOUNDED 1909. INCORPORATED 1915

Officers for 1944

J. J. CHRISTENSEN, President (1 yr.), University Farm, St. Paul 8, Minnesota
J. B. KENDRICK, Vice-President (1 yr.), University Farm, Davis, California
C. C. ALLISON, Secretary (3 yr. term expires 1944), Ohio State University, Columbus 10, Ohio
RALPH M. CALDWELL, Treasurer (3 yr. term expires 1946), A.E.S. Bldg., Purdue University, Lafayette, Indiana
HELEN HART, Editor-in-Chief of Phytopathology (term expires 1945), University Farm, St. Paul, Minnesota

Councilors for 1944

R. S. KIRBY (term expires 1945), Pennsylvania State College, State College, Pennsylvania
J. G. LEACH (term expires 1944), University of West Virginia, Morgantown, West Virginia
J. C. WALKER (term expires 1944), University of Wisconsin, Madison, Wisconsin
E. E. WILSON (Pacific Division), University Farm, Davis, California
A. G. PLAKIDAS (Southern Division), Louisiana State University, Baton Rouge, Louisiana
O. C. BOYD (New England Division), Massachusetts State College, Amherst, Massachusetts

For list of representatives and standing committees see pp. 413–414 of the May number.

BACTERIAL WILT OF TOMATO CAUSED BY PHYTOMONAS SOLANACEARUM[1]

EDWARD K. VAUGHAN[2]

(Accepted for publication August 16, 1943)

INTRODUCTION

Each season millions of tomato seedlings are shipped from the South to the Middle Atlantic and Middle Western States. Occasional fields set with these plants suffer severely from bacterial wilt infection. In some instances the disease develops soon after the plants are set out, while in other instances the plants appear to be quite healthy until as late as July or early August and then succumb to the disease. While representatives of the southern growers, the northern farmers, the canning companies, and the various experiment stations generally have agreed that the early-season infections take place in the South, there has been a great diversity of opinion concerning the infections that appeared later in the season. It seemed desirable, therefore, to determine (a) whether infected plants might appear to be perfectly healthy; (b) whether apparently healthy plants might grow vigorously for several weeks and then succumb to an infection which had taken place during the early seedling stages; and (c) whether the organism, *Phytomonas solanacearum* (E.F.S.) Bergey *et al.*, once established in Northern soils, could survive the winter and infect subsequent crops of tomatoes grown on those soils. For the benefit of the southern grower it also seemed desirable to determine the influences of soil temperature, moisture, and hydrogen-ion concentration on infection by the wilt organism.

HISTORICAL REVIEW

Bacterial wilt of solanaceous plants was first noted by Burrill (1) in 1890 and was described by Smith (10) in 1896. Since that time investigators in many countries have endeavored to control the disease. Smith (11, 12) suggested that infection could be reduced by controlling insect carriers of the disease, particularly the Colorado potato beetle, and that cultivation practices that would minimize root injury would help to prevent infection.

[1] The field data and a portion of the laboratory studies presented in this paper were conducted as a portion of cooperative investigations between the Division of Fruit and Vegetable Crops and Diseases, Bureau of Plant Industry, Soils, and Agricultural Engineering, U. S. Dept. of Agriculture, and the Georgia Coastal Plain Experiment Station, Georgia Agricultural Experiment Station, Georgia Dept. of Entomology, New Jersey Agricultural Experiment Station, and Indiana Agricultural Experiment Station. The work here reported is part of a thesis presented to the Faculty of the Graduate School of the University of Minnesota, in part-fulfillment of the requirements for the degree of Doctor of Philosophy; August, 1942.

The writer wishes to express his sincere appreciation to Dr. S. P. Doolittle and Dr. W. D. Moore for invaluable advice in planning these investigations and for assistance in carrying on many phases of the work. He also wishes to thank Dr. S. A. Wingard, Dr. G. M. Shear, Dr. I. D. Wilson, Dr. E. C. Stakman, Dr. C. J. Eide, and Betty V. Conner for assistance in preparing and editing the manuscript.

[2] Formerly Agent, Division of Fruit and Vegetable Crops and Diseases, Bureau of Plant Industry, Soils and Agricultural Engineering, Agricultural Research Administration, U. S. Dept. of Agriculture.

Nakata (6) reported that in Japan *Phytomonas solanacearum* grew only in soils with a reaction between pH 6.0 and 8.1, but Eddins (3) was able to obtain growth in culture on agars having reactions between pH 4.25 and 8.71. He found also that the organism was present in Florida soils having reactions as low as pH 4.32. He was able to control the disease, however, by treating soils with sufficient inoculated sulphur to reduce the pH to between 3.5 and 4.0.

Earle (2), van der Meer (4) and others have reported that an increase in the moisture content of infested soils results in an increased infection by *P. solanacearum*, although Nakata (6) found that there was a "detrimental water content" of the soil which would kill the organism. Meier and Link (5) found that temperatures between 77° F. and 97° F. were optimum for the development of bacterial wilt, and that the disease is inhibited at temperatures below 55° F.

METHODS AND MATERIALS

Except when other methods are mentioned, all inoculations were made by removing the plants from the soil in which they were growing, washing the soil from the roots, and then soaking the roots in a suspension of the bacteria for 5 to 15 minutes, after which the plants were reset before the roots were dry. All cultures were kept on 1.8 per cent potato-dextrose agar, and two-day-old cultures were used for transferring to test media. Most suspensions for inoculating plants were made by growing the bacteria in potato-dextrose broth for two days, although such suspensions have no apparent advantage over water suspensions of bacteria from agar cultures.

Before plants from the field or in the greenhouse were reported as being infected by *Phytomonas solanacearum* the diagnosis was confirmed by microscopic examinations; and, in many instances, also confirmed by isolating the organism and growing it in pure culture.

Source of Phytomonas solanacearum Cultures

The source of each culture used in these studies is shown in table 1. Because of the tendency, noted by Smith (12) and others, for *Phytomonas solanacearum* to die in agar cultures, the different strains were maintained in plants growing in pots of sterilized soil. Even under these conditions the organism frequently died or lost its pathogenicity, so that new cultures had to be secured from time to time. Several other cultures were used, but only those mentioned in the text are listed in table 1.

TABLE 1.—*Source of Phytomonas solanacearum cultures*

Number	Place of collection	Date of collection	Isolated by
96	New Brunswick, N. J.	August 8, 1938	E. K. Vaughan
98	Freehold, N. J.	August 17, 1938	E. W. Geigel
98x	Freehold, N. J.	August 17, 1938	E. W. Geigel
103	Moultrie, Ga.	April 28, 1939	E. K. Vaughan
151	Tifton, Ga.	February, 1942	W. D. Moore

Selection of Apparently Healthy Plants

Practically all of the tomato plants shipped to northern states from the South are inspected at various times during the growing period by state certifying agencies or by representatives of the northern plant brokers. Fields in which bacterial wilt is found are rejected, and it is doubtful whether many plants showing visible symptoms of the disease are ever shipped. Certification regulations require an interval of two years between tomato crops, and no other solanaceous crop may be grown during this period. Despite the precautions taken, a number of fields each year suffer from wilt infection, which cannot be traced to previous crops of susceptible plants.

TABLE 2.—*Development of bacterial wilt in tomato plants from infested and non-infested fields, when planted at New Brunswick, New Jersey, in 1940*

Lot No.	Number of plants set June 4	Number of plants showing symptoms of bacterial wilt		Per cent infected by wilt
		June 17	July 30	
Infested field				
1	26	1	1	3.8
2	26	1	2	7.6
3	24	6	20	83.3
4	23	7	20	86.9
5	26	0	0	0.0
6	25	2	5	20.0
7	24	14	21	87.5
8	26	4	3	11.4
Total	200	37	72	36.0
Non-infested field				
1	23	0	0	0.0
2	23	0	0	0.0
3	25	0	0	0.0
4	26	0	0	0.0
7	26	0	0	0.0
8	26	0	0	0.0
Total	199	0	0	0.0

In 1939 the plants in several badly diseased fields in New Jersey and Delaware were traced to a definite field in the southern plant-growing area. This field had been carefully checked at the time the plants were pulled, and only an occasional wilted plant was found. The northern growers reported that the plants appeared to be in excellent condition when they were received, but that the disease appeared soon after they were set out. In the spring of 1940, experimental plantings were again made in the field under suspicion and also at the Georgia Coastal Experiment Station in soil in which there had not been any bacterial wilt. Apparently healthy plants were selected from both of these plantings and shipped to New Brunswick, New Jersey, where they were set in soil the history of which showed no record of bacterial-wilt infestation. Only the strongest, healthiest plants in each lot were transplanted and each received one-half pint of water at the time of

setting. As shown in table 2, most of the lots from the field where *Phytomonas solanacearum* was known to occur suffered considerable losses because of the wilt. It is possible that the plants that developed wilt later in the season may have become infected from their diseased neighbors rather than in the southern plant field. Nevertheless, the early-season wilt, which could not have been contracted in the northern field, was of serious proportions.

These results indicate: (a) that fields in which there is a considerable amount of wilt one season should not be used for tomato-plant production the following season; (b) that apparently healthy plants may carry the causal bacteria on their roots or that there is an incipient infection that cannot be detected, and (c) that it is unsafe to use plants from fields where the disease is known to occur, even though the plants appear to be free from infection. It is interesting to note that plants from the same seed source, shipped and handled by the same people and in the same manner, but grown in the experimental plots at Tifton, Georgia, did not develop bacterial wilt at any time during the season.

OVERWINTERING OF PHYTOMONAS SOLANACEARUM IN THE SOIL

Although it was known that *Phytomonas solanacearum* lives in the soil from one season to another in the Southern States, and although Smith (11) has reported that it can be frozen in liquid air for 20 hours and yet resume its motility when thawed, it has generally been assumed that the organism cannot survive the winters as far north as New Jersey. From time to time, however, there have been outbreaks of the disease that could not be attributed to infections contracted in the South. In 1939 an outbreak occurred in the experimental plots at New Brunswick, New Jersey, in the same place where the disease was known to have been present in 1938. Since the plants were southern grown it was not possible to state definitely that the organism had lived through the winter in the soil of the experimental plot.

In order to determine whether the bacterial-wilt organism could survive the winter in the soils of New Jersey, 4 fields were selected for study. Near Riverton, a field was selected in which there had been heavy natural infestation in 1939. In fields at Old Bridge, Leesburg Prison Farm, and the New Jersey Agricultural Experiment Station, respectively, heavy infestations were induced by artificially inoculating tomato plants during the 1939 season.

The winter of 1939–40 was unusually severe, and it seemed unlikely that the bacteria would be able to survive if temperature is a limiting factor. In the spring of 1940 these 4 fields were set with Marglobe tomato plants grown from New Jersey certified seed in steam-sterilized (autoclaved) soil. Frequent observations revealed that all of the plants remained wilt-free until the soil temperature remained at or above 70° F. for several hours at a time. As soon as the soil was warm enough, wilt appeared in all 4 of the infested fields. Plants from the same source planted in autoclaved soil remained free from wilt throughout the season (Table 3).

TABLE 3.—*Survival of Phytomonas solanacearum in fields in New Jersey known to have been infested the previous season*

Location of field	Number of plants set	Per cent of plants wilted
Old Bridge	52	8
New Jersey Exp't. Station	600	20
Riverton	72	53
Leesburg Prison Farm	60	85
Autoclaved soil	50	0

In 1941 southern-grown plants set in the experimental plots at New Brunswick again were severely damaged by bacterial wilt. At the Leesburg Prison Farm tomato seed was sown in the infested soil and a large percentage of the plants produced contracted the disease. These results prove that *Phytomonas solanacearum* is capable of overwintering in the soil in most parts of New Jersey even during exceptionally severe winters and of infecting subsequent crops of susceptible plants.

SPREAD OF PHYTOMONAS SOLANACEARUM IN THE FIELD

Alternate rows of plants were inoculated in 1939 at the Leesburg Prison Farm, leaving every other row of plants uninoculated. All of the plants

Condition of plots August 1, 1939

O O O O O O O O O O O O O O O O O O O O
O O O O O O O O O O O O O O O O O O O O
O O O O O O O O O O O O O O O O O O O O
O O O O O O O O O O O O O O O O O O O O
X X X X X X X X X X X X X X X X X X X X
O O O O O O O O O O O O O O O O O O O O
X X X X X X X X X X X X X X X X X X X X
O O O O O O O O O O O O O O O O O O O O
X X X X X X X X X X X X X X X X X X X X

Condition of plots July 22, 1940, earliest date on which disease was noted

O O O O O O O O O O O O O O O O O O O O
O O O O O O O O O O O O O O O O O O O O
O O O O O O O O O O O O O O O O O O O O
O O O O O O O O O O O O O O O O O O O O
O O O O O X X X O X O O X O X O O O O O
O O O O O O O O O O O O O O O O O O O O
X O O O X O X O X O O X O O X O O O O O
X O O O O O O O O O O O O O X O O O O O
X O X O X O O X O O X O O O X X O O O O

Condition of plots August 23, 1940, date when final observations were made

O O O O O O O O O O O O O O O O O O O O
O X O O O O O O O O O O O O O O O O O O
O O O O O O O O O O O O O O O O O O O O
O X O X O X O X X X X X X O X X X X X O
X X X X X X X X X X X X X X X X X X X O
X X X X X X X X X X X X X X X X X O O O
X X X X X X X X O X O X X X X X O X O X
X X O X X X X X O X X X X X X O O O O O
X X X X X X X X X X X X O X X X X X O O

FIG. 1. Overwintering and spread of *Phytomonas solanacearum* in a field at Leesburg Prison Farm, Leesburg, New Jersey. Plants hypodermically inoculated with culture #96, July 1, 1939. O, healthy plants. X, wilted plants.

were left in the field during the winter and the soil was not cultivated or disturbed in any way. In the spring of 1940 the old plants were pulled up and replaced in each case with a healthy seedling.

When the first wilted plants were noticed in this field on July 22, most of them (19 out of 21) were found in the rows where the plants had been inoculated the previous season (Fig. 1). As the season advanced more and more wilt developed in the intervening rows until almost as many of the plants in these rows were infected as in the inoculated rows. The plants in only 3 rows were inoculated in 1939 and it is interesting to note that the plants in the three alternating rows adjacent to these were heavily invaded by the pathogen, while almost none of the plants in the rows that were at a greater distance from the inoculated ones developed the disease. The plants were set 4×4 feet apart and in most cases the organism did not attack plants more than 4 feet from the original place of infection.

At New Brunswick, where only alternate rows of plants were inoculated in 1939, but where the usual plowing and cultivating operations were carried on, there was no difference in the amount of wilt infection that appeared in the various rows in 1940. All cultivating was parallel with the rows, and tomatoes planted in an adjacent plot in both 1940 and 1941 did not become infected, although plants in the inoculated plot only a few feet distant were severely attacked in both seasons. These limited observations indicate that *Phytomonas solanacearum* does not spread rapidly from plant to plant but is dependent almost entirely on the movement of infested soil or infected plant parts or drainage water from one section of a field to another. This is further substantiated by observations made in several commercial fields over a period of 4 years. Moreover, Thomas (14) noted that wilt infections that occurred in low sections of fields in Indiana could frequently be traced to infections in higher parts of the fields. Apparently heavy rains washed the soil from the vicinity of the wilted plants to the lower parts of the field. In such instances the parts of the field adjacent to the washed areas do not become infested, the infections developing only in the flooded areas.

FACTORS INFLUENCING INFECTION BY PHYTOMONAS SOLANACEARUM

Effect of Soil Temperature

Repeated inoculation of potted plants in the greenhouse at the New Jersey Agricultural Experiment Station did not produce the disease. To determine whether this failure was due to low soil temperature, and to determine the effect of soil temperature upon the development of bacterial wilt, the following experiments were carried on at the United States Bureau of Plant Industry Station, Beltsville, Maryland.

On January 8, 1940, 3 series of 10 plants each were inoculated by dipping the roots in suspensions of bacteria or by injecting the suspensions into the stems of the tomato seedlings. The plants were then set in tanks of soil and held at temperatures of 60, 70, 80, 90, and 100 degrees F. With the excep-

tion of one series no wilt developed in any of the series of inoculated plants when the soil temperature was less than 70° F., and very little wilt developed at that temperature. In general, the amount of infection and the rate at which the disease developed, increased with a rise of temperature above 70° F. This general trend was noticeable, even in the plants inoculated hypodermically 2 inches above the surface of the soil (Fig. 2 and 3).

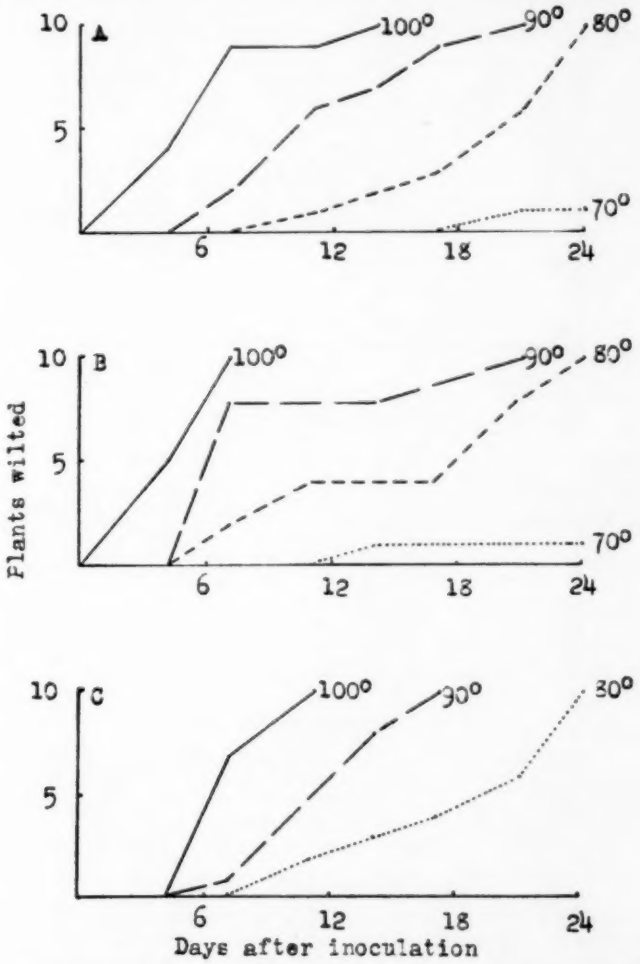

FIG. 2. Development of bacterial wilt in tomato plants inoculated by dipping the roots in a bacterial suspension and planted in soil held at various temperatures. Tests with 3 cultures. A. Culture No. 98. B. Culture No. 98–X. C. Culture No. 103. Ten plants in each treatment. No wilt developed at 60° F. with any culture and none at 70° with culture 103.

Another series of inoculations was made on February 29, 1940, by dipping the roots of tomato seedlings in a bacterial suspension. The 24 plants in each lot were set in tanks of soil kept at 70, 80, 90, 100, and 110 degrees F. Plants in soil held continuously at 110° F. did not grow well. As in the

previous experiments, however, the rate of wilt development increased with an increase in soil temperature above 70° F. (Fig. 4).

Since the critical temperature for wilt development appeared to be about 70° F., plants inoculated on March 28, 1940, by dipping the roots in a bacterial suspension were set in soil held at 60, 65, 70, 75, and 80 degrees F. While the percentage of infected plants (Fig. 5) was not so high as in the previous experiment, the results show that the amount of wilt infection increases with

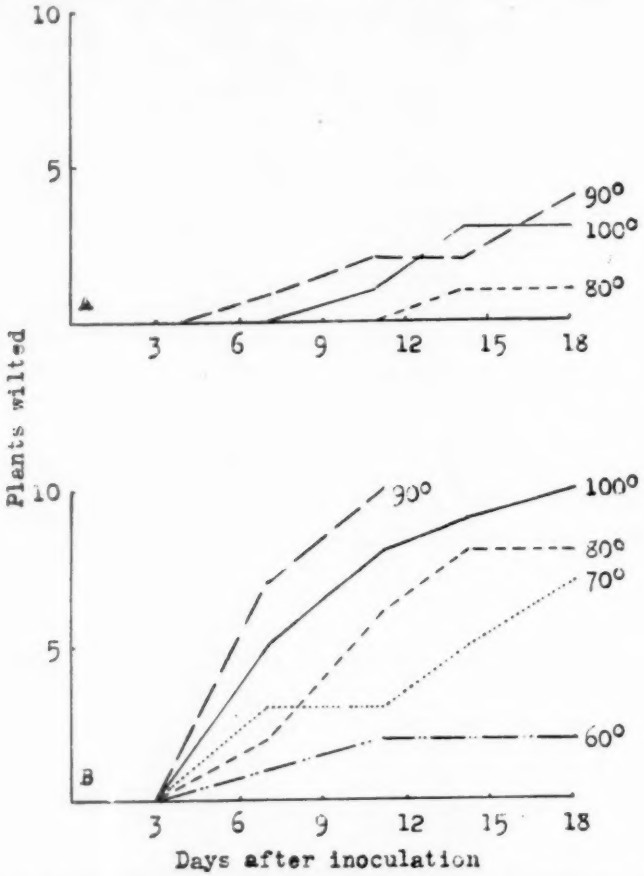

FIG. 3. Development of bacterial wilt in tomato plants inoculated hypodermically and planted in soil held at various temperatures. Test with 2 cultures. A. Culture No. 98, no wilt developed at 60° or 70° F. B. Culture No. 98–X. Ten plants in each treatment.

increased soil temperatures above 70° F. Although no wilt developed in the plants growing in soils maintained at temperatures of 60° and 65° F., a small amount developed in those growing in soils maintained at 70° F. Above that temperature the amount of wilt increased rapidly.

These results, which are similar to those of Meier and Link (4), were further substantiated by field observations. It was noted in the studies on overwintering of *Phytomonas solanacearum* in New Jersey that no wilt

appeared on plants in any of the plots in 1940 until the temperature of the soil at a depth of 6 inches, as recorded by soil thermographs, had remained above 70° F. for several days. In traveling about the State in the spring and early summer frequent readings were made with a soil thermometer. In no case was bacterial wilt observed in a locality where the soil temperature at a depth of approximately 5 inches was less than 70° Fahrenheit.

Since there was no manifestation of the disease when soil temperatures were low, it seemed desirable to determine whether infection did not take place at temperatures below 70° F., or whether infection occurred without producing the characteristic symptoms of the disease. One lot of tomato seedlings, inoculated by dipping the roots in a bacterial suspension, was

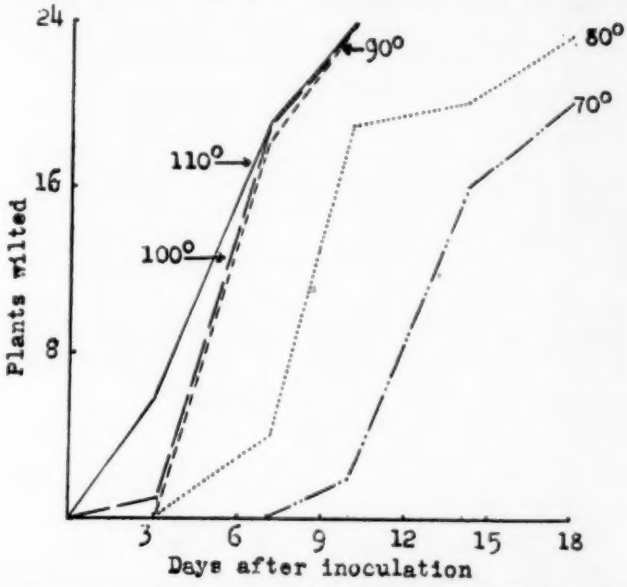

FIG. 4. Development of bacterial wilt in tomato plants inoculated by dipping the roots in a bacterial suspension and planting in soil kept at various temperatures. Culture No. 98. Twenty-four plants in each treatment.

planted in sand and kept at 75° F., a temperature at which wilt developed readily, and another lot in sand was kept at approximately 55° F., a temperature too low for the development of the disease. After 3 days both lots were removed from the sand and the roots and the bases of the stems were washed in a 1 : 3000 solution of New Improved Ceresan for 3 minutes to kill any bacteria that might be adhering to the outside of the plant.[3] After rinsing in sterile water, they were planted in steam-sterilized soil kept at a temperature of approximately 60° F. Some of the plants in each lot did not survive the Ceresan wash, but most of them soon recovered from it. As soon as the plants had recovered from the effects of the transplanting and the Ceresan wash, the temperature of the soil was raised to approximately 80°

[3] Previous tests had shown that this treatment gave 100 per cent disinfection of the root surface of plants taken directly from the soil.

F. and maintained at that point for 3 weeks. At the end of this period 53 per cent of the plants that had been kept at 55° for 3 days following the inoculation had developed the disease, compared with 78 per cent of those that had been kept at 75° for the same length of time.

These results indicate that infection can occur at a temperature of approximately 55°, although infection may take place more readily at temperatures of 70° F. and above. It seems then that plants may become infected at comparatively low temperatures, but that the symptoms of the disease do not develop unless the soil temperature remains for several days at 70° F. or higher.

Plants that had been inoculated and were severely wilted when growing in sand maintained at a temperature of approximately 80° "recovered"

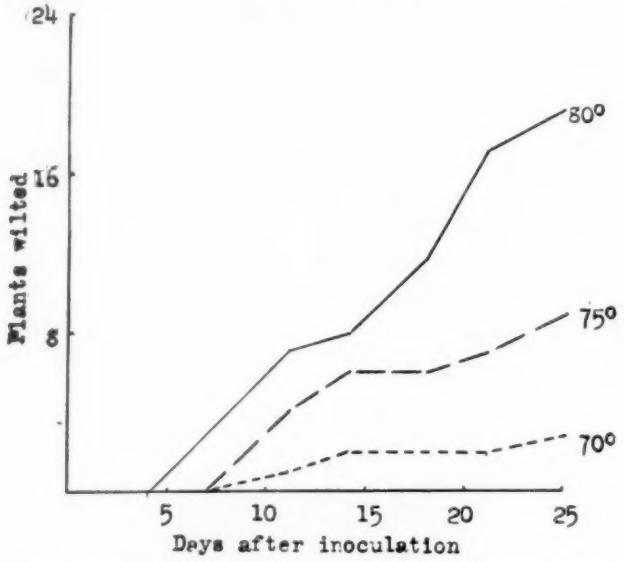

FIG. 5. Development of bacterial wilt in tomato plants inoculated by dipping the roots in a bacterial suspension and planting in soil kept at various temperatures. Culture No. 98. Twenty-four plants in each treatment. No wilt developed at 60° or 65° F.

when the temperature of the sand was lowered to approximately 55° F. for 5 days. When the temperature of the sand was again raised to 80° the plants wilted very quickly, and when the temperature was again reduced to 55° they soon "recovered." This apparent "recovery" took place from 3 to 6 times in different lots of plants before permanent wilting occurred. Infected plants in the field frequently appear to have recovered when examined in the early morning, but wilt again during the heat of the afternoon. This "recovery" may occur several times before the plant finally becomes permanently wilted. This wilting in the field probably is attributable in part to increased transpiration as the air temperature rises, but the phenomenon can be duplicated in the greenhouse by merely raising and lowering the temperature of the soil.

Effect of Soil Moisture

The literature on bacterial wilt contains many references (2, 11, 12, 14) to the fact that the disease causes greater damage in very moist soils than in drier, better drained soils. In order to gain more definite information on the effect of soil moisture upon infection by *Phytomonas solanacearum,* six

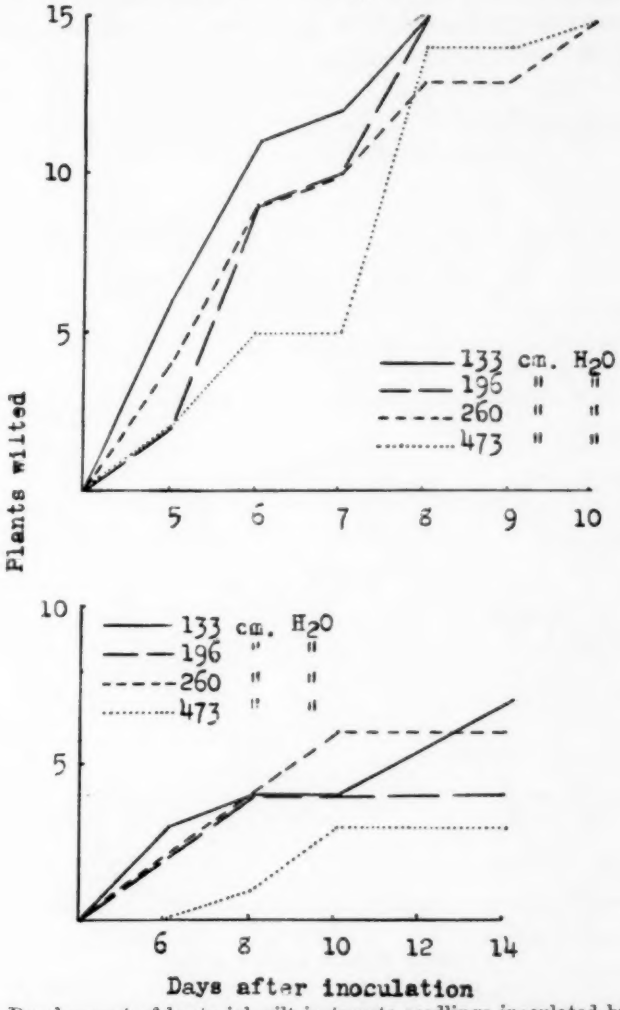

FIG. 6. Development of bacterial wilt in tomato seedlings inoculated by dipping the roots in a bacterial suspension and planting in auto-irrigator pots of soil maintained under different tensions. All pots kept at 84° F. Fifteen plants per treatment in the first test, 10 in the second.

auto-irrigation pots (8) were set up under each of the following tensions: 133, 196, 260, and 473 cm. of water. For the greater tensions, columns of mercury were used in place of water because of the limited amount of available space. Although the differences should have been greater, actual soil moisture determinations showed 21.26, 22.20, 18.48, and 16.40 per cent by

weight, respectively, at the beginning of the first inoculation test and 19.40, 14.40, 13.33, and 10.52 per cent, respectively, at the end of the second test. The pots were kept at a temperature of 84° F. and the soil in all was as uniform as thorough screening and mixing could make it. Fifteen plants in the first test and 10 in the second were set in the soil immediately after being inoculated by root dipping and when the soil had been pressed firmly around the roots the surface of the soil was covered with about one-half inch of glass wool to prevent evaporation.

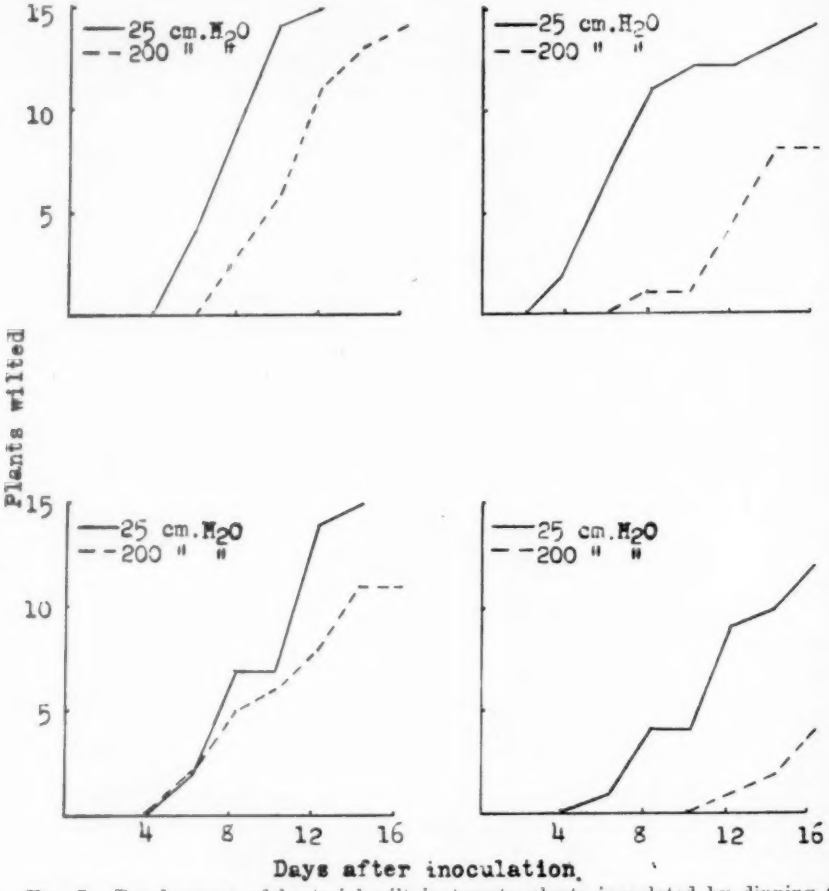

FIG. 7. Development of bacterial wilt in tomato plants inoculated by dipping the roots in a bacterial suspension and planting in auto-irrigator pots of soil maintained under different tensions. Four tests. Fifteen plants per treatment. All pots kept at 84° F.

The results of this test (Fig. 6) are interesting because they indicate that although bacterial wilt does develop more rapidly in moist soils it also may develop in comparatively dry soils, so that the ultimate damage may be equally as great in dry soils as in wet soils.

In later experiments only two tensions were used: 25 and 200 cm. of water. Determinations of the moisture content of the soil at the start of

the first inoculations were 38 and 18 per cent by weight, respectively, at the start of the third test they were 41 and 19 per cent, and at the end of the fourth test they were 36 and 16 per cent, respectively. Again the pots were maintained at a temperature of 84° F. ± 1°. The results of these tests (Fig. 7) are in general agreement with those of the first tests.

Because of the small numbers of plants in each test, it seems inadvisable to draw definite conclusions. There are indications, however, that while *Phytomonas solanacearum* is able to cause infections most readily in fairly moist soils it also can cause infections in comparatively dry soils if the percentage of moisture remains fairly constant. Where soils remain very dry, or are usually very dry except for short periods following rains, infection is not likely to occur.

Effect of Hydrogen Ion Concentration on Growth of Phytomonas solanacearum

Potato-dextrose agar was adjusted to various hydrogen-ion concentrations by the addition of HCl or NaOH to the autoclaved media, using a

TABLE 4.—*Growth of Phytomonas solanacearum on potato-dextrose agar of various hydrogen-ion concentrations*

pH of medium	Culture No. 98	Culture No. 151
4.1	–	–
4.6	–	+
4.9	+++	+++
5.4	+++	++++
6.0	++++	+++++
6.6	++++	+++++
7.1	++++	+++++
7.5	++++	+++++
8.2	++++	+++++
8.6	++++	+++++
8.9	+++	+++
9.3	++	++

– no growth
+ very faint growth
++ light growth
+++ moderate growth
++++ heavy growth
+++++ very heavy growth and the accumulation of bacteria at bottom of the streak

potentiometer to determine the reactions obtained. Bacteria from two-day-old cultures were transferred to the media and readings were made after 5 days at room temperature (70° F.). Five tubes were used in each series, and the entire experiment was run in triplicate. The results of the 3 replications were so consistent that the data from the third test (Table 4) may be taken as representative of the entire experiment.

It is obvious that *Phytomonas solanacearum* grows, at least in vitro, over a wide range of H-ion concentrations. Almost no growth occurred below pH 5 and none below pH 4.6. It made excellent growth when the pH was between 6.0 and 8.6 and developed when the pH was as high as 9.3. Its growth was not tested on media more alkaline than pH 9.3.

The cultures on media of pH 4.1 and 4.6 were transferred after 10 days to potato-dextrose agar of pH 6.8. In some cases the bacteria that had been held at pH 4.6 resumed growth on the neutral medium, indicating that not all of them had been killed by the acid medium. None of the bacteria that had been at pH 4.1 resumed growth, indicating that all of them had been killed by the acid in the medium. These results are remarkably similar to those obtained by Eddins (3), working with strains of the organism isolated from potatoes in Florida.

DISCUSSION

From the data presented in this paper it is obvious that tomato seedlings from fields in which bacterial wilt is known to be present cannot be regarded as safe for use no matter how vigorous and free from disease they may appear to be. While it may be true that infections on seedlings are severe only in localized areas in a field and, while it is not likely that a major portion of the apparently healthy plants from such field will later succumb to the disease, there is no way of knowing which plants are infected and which are not. If plants with incipient infections die soon after planting, they cannot be successfully replaced, since a healthy plant set in the space formerly occupied by a plant that died with bacterial wilt will usually contract the disease (6, 13). Thus the losses from bacterial wilt are likely to be complete and irreparable. It is necessary, therefore, to guard against setting infected plants. Since *Phytomonas solanacearum* can live over winter in the soil as far north as New Brunswick, New Jersey, it is doubly important to guard against the introduction of this organism into fields where it has not previously been known to occur.

The delay in the appearance of bacterial wilt until mid-season, after plants have grown vigorously for several weeks, may result from low soil temperatures during extended periods of cloudy weather or from earlier periods of prolonged drouth during which the soil remained very dry for a considerable period of time. By mid-season the soil temperature will have risen to 70° F. or higher, even in extended periods of cloudy weather. A few days of rainy weather would then provide the requirements for the development of the disease. It is not inconceivable that plants with incipient infections by *Phytomonas solanacearum* might remain apparently free from disease throughout the season and produce profitable crops of fruit if the proper sequence of climatic conditions prevails.

Phytomonas solanacearum usually is spread extensively in the field through a transfer of infested soil or infected plant parts by cultivating tools or by the washing caused by heavy rains. Since the rate of spread due to ordinary cultivating operations appears to be very slow, there seems to be little danger of a rapid or general spread of the disease in the field by this means.

Aside from securing disease-free plants and rotation of the crops there appears to be little that the northern grower can do to prevent or decrease the losses due to bacterial wilt. Any fertilizer which will produce vigorous,

profitable tomato plants will also provide plants in which the bacteria can thrive. Tomato plants grow best in soils having a reaction of pH 6.5 to 6.8, which is also very favorable for the development of bacterial wilt. Crop rotation, however, probably will rid the soil of the organism or at least will reduce the damage from it if non-susceptible crops are grown for several years.

The problem of the southern plant grower is more vexing. The presence of local areas of infestation in a field does not necessarily mean that the entire field is infested, but there is no way of determining the extent of the infested areas in the few days between the discovery of the disease and the time when the plants must be shipped. Since this is true the only safe course for the inspectors or buyers to follow is to reject the entire field. Should cool weather prevail throughout the brief growing period it would be impossible for the inspectors or the buyers to detect the presence of infection before the plants were shipped, and even the northern growers might not suspect presence of infection until after the plants had been set and perhaps grown for several weeks in the north.

Like the northern grower, the southern plant grower is unable to control the disease by prophylactic measures or by the use of special types of fertilizer. The margin of profit on his product per acre is so low that the application of large quantities of sulphur and lime to modify the pH of the soil sufficiently to kill the bacteria would not be profitable. Aside from crop rotation, which can reduce the incidence of bacterial wilt but will not prevent it, the only preventive practice that can be followed is a careful selection of land on which to grow tomato plants. All low, poorly-drained soils should be avoided. Fields in which the disease is known to occur should not be used. Only well-drained fields in which the disease has not been known to occur should be used, and in these fields plantings should not be made in low spots or near streams or ponds.

SUMMARY

It often is not possible to distinguish non-infected tomato plants from those having incipient *Phytomonas solanacearum* infection until they are grown in moist, warm soil.

The bacterial wilt organism can live over winter in soils at least as far north as central New Jersey.

Apparently *P. solanacearum* in the soil does not spread readily except when infested soil is moved by cultivating tools or by floodwater.

Infections may occur at soil temperatures as low as 55° F., but symptoms of bacterial wilt ordinarily do not become apparent at temperatures of 55° to 60° F. From 70° to as high as 110° F. soil temperatures, the rate of development of the disease increases with an increase in temperature.

A constant, but not necessarily a great, supply of soil moisture appears to be essential for the growth of *P. solanacearum*.

The organism is able to grow over a wide range of hydrogen-ion concen-

tration. It is killed in potato-dextrose agar cultures at pH 4, makes very little growth below pH 5, and grows best between pH 6 and pH 8.

DEPARTMENT OF BIOLOGY,
VIRGINIA POLYTECHNIC INSTITUTE,
BLACKSBURG, VIRGINIA.

LITERATURE CITED

1. BURRILL, T. J. Preliminary notes upon the rotting of potatoes. Soc. Prom. Agr. Sci. **11**: 21–22. 1890.
2. EARLE, F. S. Tomatoes. Ala. Agr. Exp. Stat. Bull. 108: 1–36. 1900.
3. EDDINS, A. H. Brown rot of Irish potatoes and its control. Fla. Agr. Exp. Stat. Tech. Bull. 299: 1–44. 1936.
4. MEER, J. H. H. VAN DER. Invloed van den vichtigheidstoestand, op de slejmziekte, veroorzookt door *Bacterium solonacearum* E.F.S. Bul. Deli Proefstation to Medan-Sumatra **39**: 47–48. 1929.
5. MEIER, F. C., and G. K. K. LINK. Potato brown rot. U.S.D.A. Cir. 281: 1–6. 1923.
6. NAKATA, K. Principles for controlling tobacco wilt. Jour. Sci. Agr. Soc. (Tokyo) **298**: 389–411. 1927.
7. N. J. Agr. Exp. Stat., Ann. Rept. **1936**: 71–72, 1936.
8. RICHARDS, L. A., and H. L. BLOOD. Some improvements on auto-irrigator apparatus. Jour. Agr. Res. [U.S.] **49**: 115–121. 1934.
9. Science and the land. Ann. Rept. N. J. Agr. Exp. Stat. **1940–1941**: 55–56. 1941.
10. SMITH, E. F. A bacterial disease of the tomato, eggplant, and potato (*Bacillus* **solanacearum** n. sp.) U.S.D.A. Div. Veg. Phys. and Path. Bull. 12: 1–28. 1908.
11. ————. The Granville tobacco wilt. U.S.D.A. Bur. Pl. Ind. Bull. 141: 17–24. 1908.
12. ————. Bacteria in relation to plant disease. **3**: 151–153, 174–219, and 220–271. Carnegie Inst. Wash. 1914.
13. ————. Bacterial diseases of plants. 170–201. W. B. Saunders and Co. (Philadelphia). 1920.
14. THOMAS, H. R. Ann. Rept. Ind. Agr. Exp. Stat. **1938–1939**: p. 53. 1939.

SOME FACTORS INFLUENCING THE SOLUBILITY OF CUPROUS OXIDE IN RELATION TO ITS TOXICITY AS A FUNGICIDE[1]

E. A. Marten and J. G. Leach

(Accepted for publication November 29, 1943)

Copper has long been recognized as one of the most effective fungicides, and in some form it has found extensive use in the control of fungus diseases of plants. Copper sulphate, partly because of its high solubility in water, was for many years the compound most frequently used. In 1919 Darnell-Smith and Ross (2) advocated the use of copper carbonate dust as a substitute for copper sulphate in treating wheat for the control of bunt. Copper carbonate, although highly insoluble in water, proved to be an effective fungicide against bunt and was much less injurious to the seed than copper sulphate. This work focussed the attention of plant pathologists on the possible fungicidal value of other copper compounds of low solubility. In recent years several so-called "insoluble" copper compounds have been found useful as fungicides.

One of the more recently exploited "insoluble" copper compounds is cuprous oxide, which was demonstrated by Horsfall (5) and others, to have considerable value as a seed protectant in the control of pre-emergency damping-off. Although highly insoluble in pure water, cuprous oxide is sufficiently soluble in moist soil to have considerable fungicidal value. Its low solubility also makes it relatively less injurious to most seeds than the more soluble copper salts. Despite this fact it has proved toxic to some seeds, and is more toxic under some conditions than others. Although its variable toxicity to seeds has limited the value of cuprous oxide as a seed protectant, it is still a promising fungicide for use as a seed protectant and for spraying and dusting.

Since the variability of cuprous oxide in both fungicidal value and in seed injury appeared to be associated with its degree of solubility under different conditions, it seemed desirable to know more about the factors influencing solubility. A review of the literature has failed to reveal any appreciable information bearing directly on the subject. Most of the work with cuprous oxide as a fungicide has been highly empirical with little attention being given to the problem of solubility in the soil in contact with germinating seeds or in close proximity to other plant tissues.

The literature is replete with references to the mechanism of the liberation of copper from a number of "insoluble" copper compounds, especially with respect to the copper in Bordeaux mixture. No attempt will be made to review all of these. Three hypotheses for the liberation of copper are quoted by McCallan (6) from the statement by Barker and Gimingham (1).

1. The copper is brought into solution by atmospheric agents—more espe-

[1] Published with the approval of the director of the West Virginia Agricultural Experiment Station as Scientific Paper No. 315.

cially by the action of carbon dioxide of the air, *i.e.*, a purely chemical explanation.

2. The leaves, on which the mixture is sprayed, exert a solvent action on the compounds, *i.e.*, an action of the host plant.

3. The fungus itself by its excretions is responsible for the production of the soluble copper by which it is finally poisoned.

McCallan and Wilcoxon (7) showed that the salts of hydroxy acids, such as malate and perhaps others, as well as of amino acids present in the spore excretions, act on Bordeaux mixture to form soluble toxic copper hydroxy and copper amino salts. While the foregoing hypotheses were intended especially to apply to Bordeaux mixture, the same might be applied with appropriate modifications to cuprous oxide when used as a dust in seed treatment. Thus, the carbon dioxide of the soil atmosphere might exert a dissolving action, or the roots of the plant could give off products with a solubilizing effect, and lastly the fungus, in intimate contact with the dust-covered seed, might excrete a solvent for the liberation of sufficient copper by which the fungus is poisoned or inhibited.

The explanation of the mechanism of solution of cuprous oxide when used in seed treatment is made more complicated by the fact that the compound comes in contact with the various constituents of the soil, which in itself is a heterogeneous mixture, and varies in composition from point to point and from soil to soil. Thus the chemical agents exerting an important solvent action at one place might be relatively unimportant at another.

In the course of certain work concerned with another phase of this problem it was observed that a sterile neutral medium containing peptone and yellow cuprous oxide became intensely blue after standing for a short time in the incubator. Since the effect was so striking, and as it offered a clue to the mechanism of solution of cuprous oxide and a plausible explanation for the immediate toxic effects that the compound shows as a fungicide in preventing damping-off, this phenomenon was further investigated. Particular emphasis was placed upon the importance of the nitrogenous products of protein decomposition in the solution of yellow cuprous oxide when used in seed treatment.

ANALYTICAL METHODS

Tests for the solubility of yellow cuprous oxide were made using water that had been doubly distilled from all-glass equipment. One hundred ml. of this water was put into a Pyrex flask together with 2 g. of yellow cuprous oxide, placed on a shaking machine, and agitated for 10 minutes each hour for one week. The oxide was then allowed to settle, whereupon the supernatant liquid was centrifuged 3 times for 15 minutes each at 3000 r.p.m. This triply centrifuged water was tested (method 2) and found to contain from 0.6 to 0.8 p.p.m. of copper. When ordinary laboratory distilled water was used and treated in the same way, as much as 1 to 2 p.p.m. of copper was found present. Methods used for the estimation of copper:

1. Whenever the amount of copper in solution was between 100 and 800 p.p.m. the solutions were analyzed directly, but if they exceeded the upper-limit they were diluted so as to fall within this range. Four ml. of the copper solution was mixed with 1 ml. of concentrated ammonium hydroxide. The intensity of color of the resulting blue solution was determined with a Fischer Electrophotometer, using the A. filter. Reference of the reading to a standard curve gave the amount of copper in solution. The solutions used in constructing the standard curve were prepared by dissolving weighed amounts of pure copper sulphate ($CuSO_4 \cdot 5H_2O$) in water containing 1.0 per cent and 0.1 per cent of glycine.

2. The principle of the Biazzo method as modified by Elvehjem and Lindow (3) was employed for solutions containing less than 100 p.p.m. of copper. The neutral solution of copper was diluted so as to contain from 2 to 20 p.p.m. of copper. Ten ml. of this solution was placed in test tubes of uniform diameter. The following reagents were added, shaking after each addition: 1 ml. of glacial acetic acid; 1 ml. of 10 per cent potassium thiocyanate solution; 5 drops of freshly distilled pyridine, and 5 ml. of chloroform. The tubes were allowed to stand until the chloroform layer became clear. The yellow color of the chloroform layer was compared with that obtained in a set of standard tubes prepared in the same way. When the amount of copper was less than 2 p.p.m. the color was determined with a Keufel and Esser color analyzer using the wave length 450 m/w to obtain the readings. The color was compared with that of a standard copper solution similarly treated.

3. Qualitative tests for copper were made by adding 3 to 5 drops of a 5 per cent solution of a-benzoin oxime in 95 per cent alcohol to 5 ml. of the sample solution. The formation of a yellow color showed the presence of copper.

Influence of Certain Nitrogenous Compounds on Solubility of Cuprous Oxide

In order to determine the solvent action of nitrogenous substances on cuprous oxide gelatin, peptone, glycine, aspartic acid, asparagine, arginine, and cystine were tested. The materials were dissolved in distilled water in the concentrations indicated in table 1. An excess of yellow cuprous oxide was added to the flasks. These were then stoppered and placed on a shaking machine, which agitated the liquids for 10 minutes every hour during 7 days at a temperature of 25° C. The amounts of copper brought into solution by these materials are shown in table 1.

The data show that the hydrolyzed protein and pure amino acids, which were soluble in water, dissolved the cuprous oxide readily, and that this was accomplished by very dilute solutions of these chemicals. By way of contrast, the relatively unhydrolyzed protein gelatin, when used in the same concentrations, produced comparatively little effect.

In order to determine the effect of the products of bacterial decomposition of gelatin the following experiment was performed. Coarse white

TABLE 1.—*The solvent action of certain protein derivatives on cuprous oxide*

Material	Concentration	Copper in solution
	Grams per 100 cc.	*p.p.m.*
Blank	.....	0.6 to 0.8
Gelatin	0.01	2
"	0.10	10
"	1.00	125
Peptone	0.01	2
"	0.10	100
"	1.00	925
Glycine	0.001	7
"	0.01	50
"	0.10	550
"	1.00	2200
Aspartic acid	0.10	370
Asparagine	0.10	220
Arginine	0.10	185
Cystine	Saturated sol'n.	45

sand was mixed with two per cent of gelatin. Fifteen grams of this mixture was placed in a 250-ml. flask and sterilized. Four ml. of a water suspension of bacteria was mixed with the sand, and the cultures were incubated at 27° C. for 1 week. Then 0.1 g. of yellow cuprous oxide was added to each flask and mixed with the sand. This was allowed to stand for 1 day at 27° C. after which 5 ml. of water was added and the whole allowed to stand with frequent shaking for one hour. The liquid was poured from the sand and centrifuged 3 times for 15 minutes each at 3000 r.p.m. The clear supernatant liquid was tested qualitatively for copper, using a 5 per cent alcoholic solution of a-benzoin oxime as a test reagent. The results are given in table 2. In those flasks inoculated with proteolytic bacteria a greater amount of copper was brought into solution than in those inoculated with the non-proteolytic *Escherichia coli*, or in the control flasks.

Further evidence that the products resulting from the bacterial decomposition of proteins are factors in the solution of yellow cuprous oxide, was obtained by placing cubes of hard-boiled egg white weighing 1 g. in 10 ml. of sterile distilled water in flasks inoculated with a water suspension of bacteria and incubated for 3 days at 37° C. After this, the solutions were filtered through paper and yellow cuprous oxide added to the filtrate in a flask. A drop of toluene was added to each flask to prevent further growth

TABLE 2.—*The relative solvent action of the products of bacterial decomposition of gelatin*

Bacteria used	Solvent index[a]
None	1
Escherichia coli	1
Bacillus mesentericus	6
Bacillus subtilis	5
Proteus vulgaris	3
Bacteria from garden soil	4

[a] Based on the relative intensity of the color produced by the a-benzoin-oxime test.

of the organisms. After standing for 24 hours at 25° C. the solutions were cleared by centrifuging them 3 times for 15 minutes each at 3000 r.p.m. The clear solutions were tested for copper.

Eight p.p.m. of copper were found in the solution containing egg white alone. When the non-proteolytic bacterium *Escherichia coli* was added, only a slight increase of copper in solution was noted, but when proteolytic species were used the amount of copper was increased as follows: *Proteus vulgaris,* 380 p.p.m., and *Bacillus subtilis,* 670 p.p.m.

The results of this experiment show again that the products of protein decomposition are active agents in dissolving cuprous oxide. A method employing an entirely different principle demonstrated the solubility of cuprous oxide and a probable mechanism by which copper becomes effective as a fungicide when employed in seed treatment. A 4 per cent solution of the best grade of agar-agar was drawn into a glass tube with an internal diameter of 0.25 in. and allowed to solidify. The cylinder of agar was pushed from the tube into a beaker of distilled water, in which the agar was dialyzed for 24 hours with frequent changes of the water. Thereafter, the

TABLE 3.—*The solubility of cuprous oxide in several substrates as indicated by the absorption of copper by agar cylinders*

Substrate	Solvent index[a]
Control agar cylinder	0
Water saturated with Cu_2O. Excess of Cu_2O on bottom of flask	1
White sand	2
Potting soil	3
Sand moistened with 0.1% glycine water	4
Sand moistened with 1.0% glycine water	6

[a] Based on relative intensity of the color produced by the a-benzoin-oxime test.

cylinder was cut into 1-in.-long sections, the free water removed between pieces of filter paper, rolled in finely powdered dry copper oxide and then imbedded in sand or other test material, moistened and allowed to stand for 48 hours. The cylinders were then removed, washed in distilled water to remove sand, dirt, and excess copper oxide, dried between filter paper, and split lengthwise, thus giving 2 sections, with a broad surface throughout the length of the cylinder. The sections were flooded with a 5 per cent alcoholic solution of a-benzoin oxime, whereupon a greenish yellow coloration appeared, the intensity of which indicated the amount of copper absorbed by the cylinder. The data show that the composition of the soil and soil-water affect the liberation of copper (Table 3). Only a small amount of copper is put into solution by distilled water itself, however, a greater amount is liberated when organic matter of nitrogenous composition is present. This is especially true if the nitrogenous organic matter is an amino acid.

In order for a fungicide to be effective in preventing damping-off, it must go into solution rapidly enough to affect the pathogen before it has time to attack the host plant. In the case of rapidly germinating seeds, copper must be liberated shortly after the seeds have been planted, for the protec-

tive influence of the fungicide must exert itself as soon as the seed begins to sprout. In the short time required between planting and germination, some mechanism other than mere solution by water alone must be active to liberate sufficient copper.

Since the products of protein hydrolysis dissolved cuprous oxide, the following experiment was conducted to determine the speed with which the copper was released. One-half gram of cuprous oxide was added to 500 ml. of 0.1 and 0.01 per cent solutions of glycine at 25° C. and the mixture shaken frequently during the test period. At specified times samples were removed, clarified by centrifuging, and the amount of copper in solution determined. The results are given in table 4 from which it is evident that large amounts of copper are put into solution within a period of time, sufficiently short to make the toxic element available in effective concentration around the seed upon its emergence.

TABLE 4.—*The rate of solution of cuprous oxide in glycine solutions*

0.1 per cent glycine		0.01 per cent glycine	
Time of action	Copper	Time of action	Copper
hr.	*p.p.m.*	*hr.*	*p.p.m.*
0	0	0	0
½	75	½	15
3	105	1½	25
17	210	3½	50
24	280	5	55
		24	75

The results of these experiments show that the nitrogenous products of protein decomposition are active in the rapid solution of cuprous oxide. It is not suggested that this is the only or main mechanism of solution, but it does seem possible that this factor may play an important role in the action of cuprous oxide in controlling damping-off. The oxide-coated seeds lie for some time in moistened soil. The latter contains a mixture of protein in various stages of decomposition. These highly active reagents quickly liberate sufficient copper to protect the seedlings from attack by the fungus. It is also probable that this solvent action, by releasing too much copper, may account for the seed injury sometimes obtained with copper oxide.

The Toxicity of Copper Brought into Solution by Certain Nitrogenous Compounds

Increased solubility does not necessarily imply correspondingly greater toxicity. Heuberger and Horsfall (4) have shown that materials containing considerable amounts of protein, such as derris powder, pyrethrum powder, soya flour, and alfalfa meal, reduced the fungicidal value of copper compounds. These authors suggested that this action is most likely brought about by a reaction between the toxic copper and the protein, which reduces the amount of the toxic copper available to the spores. It seemed desirable,

therefore, to investigate the toxicity of the copper solution resulting from the action of these nitrogenous substances on cuprous oxide.

Methods and Materials. A master copper solution was prepared by dissolving a weighed amount of copper sulphate ($CuSO_4 \cdot 5H_2O$) in water containing the desired concentration of glycine or other test substances. This solution was neutralized to a pH of 6.2. Another stock solution was prepared by dissolving a weighed amount of yellow cuprous oxide in water containing either glycine or peptone and neutralizing this to a pH of 6.2. Dilutions of the master solutions (giving the desired concentration of copper), were prepared as required. Subsequent experiments showed that the copper was of the same toxicity from either master solution, indicating that

TABLE 5.—*The effect of different amounts of gelatin, peptone, and glycine on the toxicity of copper solutions as indicated by the relative growth of Pythium*

Composition of test medium	Copper	Relative growth of *Pythium*[a]
	p.p.m.	
Basal medium in water	0.0	4
" " " "	0.3	4
" " " "	0.5	–
" " " " + 0.1% gelatin	1.0	4
" " " " " " "	2.0	–
" " " " " " peptone	5.0	4
" " " " " " "	7.0	–
" " " " " " glycine	7.0	4
" " " " " " "	10.0	2
" " " " " " "	12.0	–
" " " " " 1.0% gelatin	5.0	4
" " " " " " "	7.0	–
" " " " " " peptone	60.0	4
" " " " " " "	80.0	2
" " " " " " "	100.0	–
" " " " " " glycine	200.0	4
" " " " " " "	225.0	3
" " " " " " "	250.0	–

[a] 4 = Good growth.
– = No growth.

toxicity is a function of the copper ion rather than the copper compound from which it is obtained.

The basic nutrient consisted of a solution prepared by adding 1 g. of potato-dextrose agar (Difco) and 0.1 g. of peptone to 20 ml. of distilled water. This was allowed to stand with frequent shaking for 2 hours at room temperature, then filtered. The solution was sterilized and 0.2 ml. of it was added to 10 ml. of the sterile copper-containing test solution. This procedure was employed in order to reduce decomposition of the nutrients to a minimum.

The test organism was a laboratory strain of *Pythium debaryanum*, which caused active damping-off of spinach and other seedlings. The fungus was grown in the basic medium for 5 days. Small pieces of mycelium were added to 10 ml. of test solution contained in a 250-ml. Erlenmeyer flask.

The flasks were incubated at 27° C. for 5 to 7 days and then examined for the amount of growth.

Experimental Results. The effect of gelatin, peptone, and glycine upon the toxicity of copper to *Pythium* is recorded in tables 5 and 6. In the experiment reported in table 5 the growth of *Pythium* was estimated. In the one reported in table 6 growth was measured by determining the dry weight of mycelium formed. These results show that the threshold of toxicity was between 0.3 and 0.5 p.p.m. in a solution containing only the basic nutrients. Upon the addition of gelatin, peptone, and glycine, the tolerance for copper increased. The reduction of toxicity varied with the kind and amount of

TABLE 6.—*The effect of different amounts of gelatin, peptone, and glycine on the toxicity of copper solutions as indicated by the dry weight of mycelium formed by Pythium*

Composition of test medium	Copper	Dry wt. of mycelium
	p.p.m.	*mg.*
Basal medium in water	0.0	6.5
" " " "	0.3	5.1
" " " " + 0.1% gelatin	0.0	15.5
" " " " " " "	0.5	7.8
" " " " " " "	1.0	9.1
" " " " " 1.0% "	0.0	15.1
" " " " " " "	3.0	13.5
" " " " " " "	5.0	14.1
" " " " " 0.1% peptone	0.0	13.1
" " " " " " "	1.0	9.9
" " " " " " "	3.0	7.7
" " " " " 1.0% "	0.0	26.0
" " " " " " "	40.0	18.3
" " " " " " "	60.0	20.1
" " " " " 0.1% glycine	0.0	11.8
" " " " " " "	3.0	7.5
" " " " " " "	5.0	6.5
" " " " " 1.0% "	0.0	11.1
" " " " " " "	150.0	5.9
" " " " " " "	200.0	6.9

nitrogenous substance added to the medium. The depression of the toxic action was least pronounced in the case of gelatin, a slightly hydrolyzed protein, and progressively more in the case of peptone and glycine, both of which result from the extensive hydrolysis of proteins. Furthermore, the neutralizing influence increased with an increase in the concentration of test material.

An estimate of the toxicity-depressing effect can be obtained from table 7. Whereas the threshold of toxicity in the basal medium alone was between 0.3 and 0.5 p.p.m. of copper, the addition of 0.5 per cent of glycine permitted the growth of the fungus in only slightly diminished amounts until a concentration of 100 to 110 p.p.m. was reached, after which growth ceased.

In order to determine the amount of glycine necessary to neutralize the toxicity of a given amount of copper an experiment was conducted wherein the amount of copper in solution was kept constant and the amount of

TABLE 7.—*The effect of 0.5 per cent glycine on the toxicity of different concentrations of copper as indicated by the dry weight of mycelium formed by Pythium*

Composition of test medium	Copper	Dry wt. of mycelium[a]
	p.p.m.	*mg.*
Basal medium	0.0	7.0
" "	0.3	7.2
" "	0.5	1.2
" " + 0.5% glycine	0.0	10.8
" " " " "	60.0	9.3
" " " " "	80.0	8.4
" " " " "	100.0	9.5
" " " " "	110.0	0.7

[a] The weight of the combined mycelium from three replicate flasks.

glycine progressively increased. The results are given in table 8. The data show that from 0.5 to 0.6 per cent of glycine were required to overcome the toxicity of 100 p.p.m. of copper.

It has been shown that dilute solutions of glycine dissolved many times as much cuprous oxide as was dissolved by distilled water. The question arises as to what extent a given amount of glycine would reduce the toxicity of the copper that it put into solution. The data in table 9 show the relationship between the dissolving power of glycine and the toxicity of the copper-glycine complex. From the table it may be seen that the copper-glycine complex is toxic in concentrations far below the total amount of copper that is put into solution by glycine. In order to be toxic approximately 500 times as much copper is required when 1 per cent of glycine is present as is necessary in the basal medium alone.

These results confirm the conclusions of Heuberger and Horsfall (4) that nitrogenous compounds may reduce the fungicidal value of copper compounds. This is true, however, only when an excess of the materials is present. Smaller quantities may actually increase the toxicity by increasing the solubility of the copper compound. It is not known definitely in what way excess nitrogenous compounds decrease the toxicity of the copper,

TABLE 8.—*The amount of glycine required to neutralize the toxicity of a 100 p.p.m. solution of copper as indicated by the relative growth of Pythium*

Glycine	Copper	Relative growth[a]
%	*p.p.m.*	
0.0	0.0	4
0.1	100.0	–
0.2	"	–
0.3	"	–
0.4	"	–
0.5	"	–
0.6	"	3
0.7	"	4

[a] 4 = Good growth.
– = No growth.

but it is probably due to reduced ionization of new compounds formed by the interaction of the two materials.

Nikitin and Anderson (8) studied the effect of various protein-containing supplements on the adherence and activation of fixed copper fungicides. Their results showed that the magnitude and tenacity of the spray residue are considerably increased by supplements containing proteins. Furthermore, these supplements exert a dissolving action on fixed copper fungicides. However, no studies were reported on the toxicity of the copper put into solution by the proteins. Since these findings (with respect to the solution of copper) were in line with those of the writers when using cuprous oxide, a study was made of the dissolving effect of soya-bean flour under specific conditions and the toxicity of the resulting solution.

One gram of finely ground soya-bean flour was mixed with 100 ml. of distilled water. This was allowed to stand in the ice box with frequent

TABLE 9.—*The relationship between the dissolving power of glycine and the toxicity of the copper-glycine complex*

Concentration of glycine	Copper dissolved	Threshold of toxicity[a]
%	*p.p.m.*	Cu *p.p.m.*
0.000	0.8	0.3 to 0.5
0.001	7.0	0.3 to 0.5
0.010	50.0	0.5 to 0.6
0.100	550.0	0.8 to 1.0
0.500	1800.0	90 to 100
1.000	3200.0	225 to 250

[a] "Threshold of toxicity" means the smallest amount of copper required to inhibit the growth of *Pythium*.

shaking for 48 hours. Thereafter, the liquid was filtered through paper and further clarified by centrifuging. An excess of washed Bordeaux-mixture precipitate was added to the soya-bean extract and allowed to stand with frequent shaking at 20° C. for 48 hours. The mixture was clarified by centrifuging and filtering. A control, using water and Bordeaux mixture, was treated in the same way. The resulting solutions were tested for copper and for their toxicity toward *Pythium debaryanum*.

Copper was not detected in the water control, while the soya-bean-extract sample contained 220 p.p.m. of copper. The control solution was not toxic, whereas the soya-bean-extract preparation was completely toxic in a dilution of 1:400, but permitted growth in a dilution of 1:450. The amount of copper in the highest dilution of soya-bean-copper solution was approximately 0.5 p.p.m., thereby indicating that the toxicity of this soluble copper complex is of essentially the same magnitude as that of the copper from inorganic sources, such as copper sulphate.

Experiments were conducted to determine the effect of soya-bean flour on the solution of yellow cuprous oxide and to test the toxicity of the copper solutions so obtained. A 1 per cent suspension of soya-bean meal was dispersed in 100 ml. of water. An excess of yellow cuprous oxide was added

to the mixture, which was allowed to stand, with frequent shaking, for 4 days at 10° C. The resulting solution was clarified by filtering and centrifuging. Analysis of this solution showed that it contained 125 p.p.m. of copper. *Pythium debaryanum* did not grow in this solution when tested in the usual way. Furthermore, the solution, upon dilution with distilled water, prevented growth of the fungus at a concentration of 0.6 p.p.m. of copper. When fresh soybean meal was added to the clarified solution a reduction in toxicity occurred. The threshold of toxicity rose to 2 to 3 p.p.m. when 0.1 per cent of meal was added, and complete loss of toxicity resulted when 1 per cent was used.

Thus it was shown that soya-bean flour is capable of activating the copper of cuprous oxide in far greater quantities than the minimum necessary for a toxic concentration. Some of the liberated copper is unquestionably bound by the soya-bean flour, however, the amount liberated is greater than the minimum amount necessary for the inhibition of growth of the fungus, so long as an excess of active soya-bean is not present. The addition of an excess of soya-bean flour to a solution containing copper reduces its toxicity. This fact should be kept in mind when using protein-containing supplements with copper fungicides, as suggested by Nikitin and Anderson (8).

SUMMARY

A study has been made of certain factors influencing the solubility and toxicity of cuprous oxide using *Pythium debaryanum* as a test organism.

Growth of *P. debaryanum* in a standard liquid medium was inhibited by cuprous oxide at a concentration of 0.3 to 0.5 p.p.m. of copper in solution. As much as 0.6 to 0.8 p.p.m. of copper was brought into solution by prolonged action of doubly distilled water. Ordinary laboratory distilled water dissolved from 1 to 2 p.p.m.

The solubility of cuprous oxide is greatly increased by glycine and other nitrogenous products of protein decomposition. As much as 2200 p.p.m. was liberated by the addition of 1 per cent glycine. The copper liberated by these nitrogenous compounds is equally as toxic as the copper dissolved in distilled water provided no excess of nitrogenous compounds is present. However, the threshold of toxicity in p.p.m. is greatly increased when an excess of these compounds is present, 1 per cent glycine raising the toxicity threshold from 0.3 p.p.m. to 225 p.p.m.

Soya-bean flour increased the solubility of cuprous oxide, a 1 per cent suspension dissolving 125 p.p.m. The copper dissolved by soya-bean flour inhibited growth of *P. debaryanum* when diluted to 0.6 p.p.m. but when 0.1 per cent of soybean meal was added to the diluted solution the threshold of toxicity was increased to 2 to 3 p.p.m. and toxicity was completely lost by the addition of 1 per cent soya-bean flour.

Because nitrogenous products of bacterial decomposition are known to be present in varying amounts in arable soils, it is very probable that they influence the solubility of cuprous oxide used as a seed protectant. They

may account for some of the variability in seed protection and seed injury experienced with cuprous oxide.

In as much as the influence of nitrogenous compounds on the toxicity of copper fungicides depends, not only upon the nature of the compound, but also on the ratio between the two substances, caution must be used in evaluating supplements containing proteins. Excess of protein supplements may decrease the toxicity of copper fungicides, while smaller amounts may, by increasing the solubility of copper compounds, increase their fungicidal value.

AGRICULTURAL EXPERIMENT STATION,
MORGANTOWN, WEST VIRGINIA.

LITERATURE CITED

1. BARKER, B. T. P., and C. T. GIMINGHAM. The fungicidal action of bordeaux mixtures. Jour. Agr. Sci. 4(1911–12): 76–94. 1913.
2. DARNELL-SMITH, G. P., and H. ROSS. A dry method of treating seed wheat for bunt. Agr. Gaz. N. S. Wales. 30: 685–692. 1919.
3. ELVEHJEM, C. A., and C. W. LINDOW. The determination of copper in biological materials. Jour. Biol. Chem. 81: 435–443. 1929.
4. HEUBERGER, J. S., and J. G. HORSFALL. Reduction in fungicidal value of copper compounds by organic materials. Phytopath. 32: 370–378. 1942.
5. HORSFALL, JAMES G. Red oxide of copper as a dust fungicide for combating damping-off by seed treatment. New York State Agricultural Experiment Station, Geneva, N. Y., Bulletin No. 615. 1932.
6. MCCALLAN, S. E. A. The solvent action of spore excretions and other agencies on protective copper fungicides. Cornell University Agricultural Experiment Station. Memoir 128, May, 1930.
7. ————, and FRANK WILCOXON. The action of fungous spores on bordeaux mixture. Contributions from Boyce Thompson Institute 8(2): 151–165. 1936.
8. NIKITIN, A. A., and E. G. ANDERSON. The adherence of fixed copper fungicides as influenced by spray supplements. Phytopath. 32: 953–964. 1942.

VARIATION IN SYMPTOMS PRODUCED BY ISOLATES OF PHYTOMONAS MEDICAGINIS VAR. PHASEOLICOLA

J. H. Jensen and J. E. Livingston[1]

(Accepted for publication January 20, 1944)

INTRODUCTION

In greenhouse halo-blight-resistance tests, inoculated bean leaves were observed on which necrotic halo-less lesions appeared, occasionally in addition to lesions with characteristic halos. Since the inoculations were carried out with an intentional mixture of several isolates and, since the 2 types of lesions sometimes appeared side by side on the same leaf, it seemed probable that the atypical ones might have resulted from infection with certain strains of the bacterium rather than because of temperature or varietal reaction.

It has been noted by several investigators that symptom variations in halo blight occur. Burkholder (1) noted that halos are never produced around the small, necrotic leaf lesions when infection takes place in the hot days of July or August. Goss (3) demonstrated that only halo-less lesions appeared at 28° and 32° C. on leaves of Red Kidney and U.S. No. 5 Refugee, whereas typical halos surrounded lesions on leaves of plants of these two varieties held at 16°, 22°, and 24° C. More recently (4) it was shown that in certain varieties, Mexican Red, Schwert, and others, only small, necrotic halo-less lesions were produced under a wide range of temperatures (16°–28°) and at all ages of growth, whereas Red Kidney and Bountiful plants inoculated at the same time with the same inoculum produced lesions with characteristic halos at 16° and 22°.

In preliminary tests with 6 isolates it was observed that at least 2 isolates regularly produced some leaf lesions that lacked the characteristic halos. Further tests with isolates from several parts of the United States, as well as other isolates from Nebraska, showed conclusively that in addition to the variation in halo production a rather wide range of symptoms is produced by isolates of *Phytomonas medicaginis* var. *phaseolicola* (Burkh.). A description of the range of these symptoms with various inoculation methods on Red Kidney beans and a summary of the physiological studies with these isolates on artificial media are presented in this paper.

MATERIALS AND METHODS

The 13 isolates of *Phytomonas medicaginis* var. *phaseolicola* Burkh. used in these studies were obtained from naturally infected field-grown beans from various localities including Wisconsin, Montana, Louisiana, and Nebraska. Since the isolates had been in culture on artificial media for lengths of time varying from several weeks to several years, it seemed desirable to attempt to eliminate the possible variations due to age-of-culture effect.

[1] Published with the approval of the Director as Paper No. 349 Journal Series, Nebraska Agricultural Experiment Station.

Accordingly, all isolates were inoculated by needle puncture into stems of young, healthy Red Kidney plants whence they were isolated after a period of several weeks. All studies reported here were carried out with the reisolates, whose pathogenic characteristics were identical with the original isolates.

Single-cell isolations of the bacteria were not attempted. All stock cultures were obtained from isolated single colonies on plates poured with high dilutions. That striking differences were obtained by cultures from isolated single colonies indicates that similar differences might be found when working with colonies grown from single bacterial cells.

Stock cultures were maintained on bean-decoction agar consisting of the boiled decoction of 200 g. of bean leaves made up to a liter of 2 per cent agar. Physiological studies were carried out on media made up according to standards adopted by the Society of American Bacteriologists (2).

Plants were grown in composted soil in 5-inch porous clay pots in the greenhouse during autumn, winter, and spring months. Greenhouse temperatures were held at approximately 22° C.

Studies of the symptoms produced by the various isolates were made by using 4 inoculation techniques: leaf, stem, germinated seed, and pod inoculations. Although complete descriptions of these methods are presented in a previous publication (4), a brief description is given here. Leaf and pod inoculations were made by spraying plants with a bacterial suspension in water. Previous to inoculation, plants were held overnight in an incubation chamber at 24° C. maintained at a high humidity. Following the inoculation plants were held in the incubator for 6 or 7 hours before removal to a greenhouse bench. Leaf inoculations were made just as the third trifoliate leaves were unfolding, and pod inoculations were made during pod development. Stem inoculations were made by stabbing twice through a smear of bacteria placed on the bean stems about ½ cm. below the primary leaves. The method used in making germinated seed inoculations consisted of soaking germinated seeds for 4 hours in a dilute bacterial suspension in water. Seeds were germinated in a moist rag-doll held for 60 hours at 27°–28° C.

Except in the case of pod inoculations, these pathogenicity tests were conducted by making inoculations with all isolates at the same time. The incubation chambers were too small to accommodate at one time all of the nearly mature plants used in pod inoculation; therefore, inoculations were made in 2 sections carried out on successive days.

Following the first complete series of tests, conducted with bacteria isolated from inoculated Red Kidney plant stems, all isolates were transferred to agar slants which, after a day's growth of the bacteria, were covered with sterile mineral oil (5). The 4 types of inoculation tests were repeated a number of times over a 4-year period with isolates stored under oil for periods varying from several months to several years. Such extensive tests failed to show any major changes in pathogenicity or in growth characteristics as a result of storage. In some tests isolates that had been stored under

oil were compared with the same isolates maintained by routine laboratory transfer, and no changes ascribable to either treatment were shown.

EXPERIMENTAL RESULTS

Leaf Inoculations

Healthy, young Red Kidney plants were sprayed with water suspensions of the bacteria. Five plants were used for each isolate. The first symptoms appeared within 5 days on plants inoculated with certain isolates; whereas, in other cases the first symptoms appeared 1 to 3 days later. Final readings were not made until 15 to 20 days after inoculation. Table 1 presents

TABLE 1.—*Some differential symptoms produced on Red Kidney beans by 13 isolates of Phytomonas medicaginis var. phaseolicola in four methods of inoculation. Isolates are presented in three groups, the spaces separating them on basis of pathogenicity*

Isolate number	Leaf inoculation		Stem inoculation				Germinated seed inoculation[c]	Pod inoculation
	Lesion type	Systemic infection	Stunting	Vein-clearing	Wilting	Mottling	Vein-clearing primary leaf	Lesions
2076[a]	Halo	++[b]	+++	+++	+++	0	+++	Typical
2080	Halo	++	+++	+++	+++	0	+++	Typical
2082	Mixed	0	+++	+++	+++	0	+++	Typical
2071	Mixed	0	+++	+++	+++	0	+++	Typical
2072	Mixed	0	++	++	++	0	++	Typical
2074	Mixed	0	++	++	++	0	0	Small
2077	Mixed	0	++	+	++	++	0	Typical
2078	Mixed	0	++	+	++	0	0	Typical
2081	Mixed	0	++	+	++	0	0	Typical
2073	Halo-less	0	++	+	+	++	0	Small
2079	Halo-less	0	+	0	0	++	0	Small—few
2083	Halo-less	0	+	0	0	++	0	Small—few
2075	Halo-less	0	0	0	0	0	0	Small

[a] The authors are indebted to John McLean, Colorado State College, Fort Collins, Colo.; Lee Person, Louisiana State University, Baton Rouge, La.; M. M. Afanasiev, Montana State College, Bozeman, Mont., for some of the isolates used in these tests.

[b] Symbols: +++ = severe, ++ = moderate, + = mild, 0 = did not occur.

[c] Stem cankers were produced by all isolates with germinated-seed inoculation method.

a summary of the symptoms produced by all isolates and lists them in 3 general groups or levels of pathogenicity. Large, halo lesions always appeared on plants inoculated with 4 of the isolates. Small, necrotic, halo-less lesions invariably appeared on plants inoculated with 4 other isolates. On the other hand, a mixture of halo and halo-less lesions appeared on plants inoculated with the remaining isolates. Where mixed lesion types occurred, these 2 types of lesions frequently appeared side by side on the same leaf. Figure 1, G, H, and I presents photographs of typical halo, halo-less, and mixed lesion types.

Usually, isolates producing halo or mixed lesions also produced the greatest number of lesions. An exception, however, was also observed. One

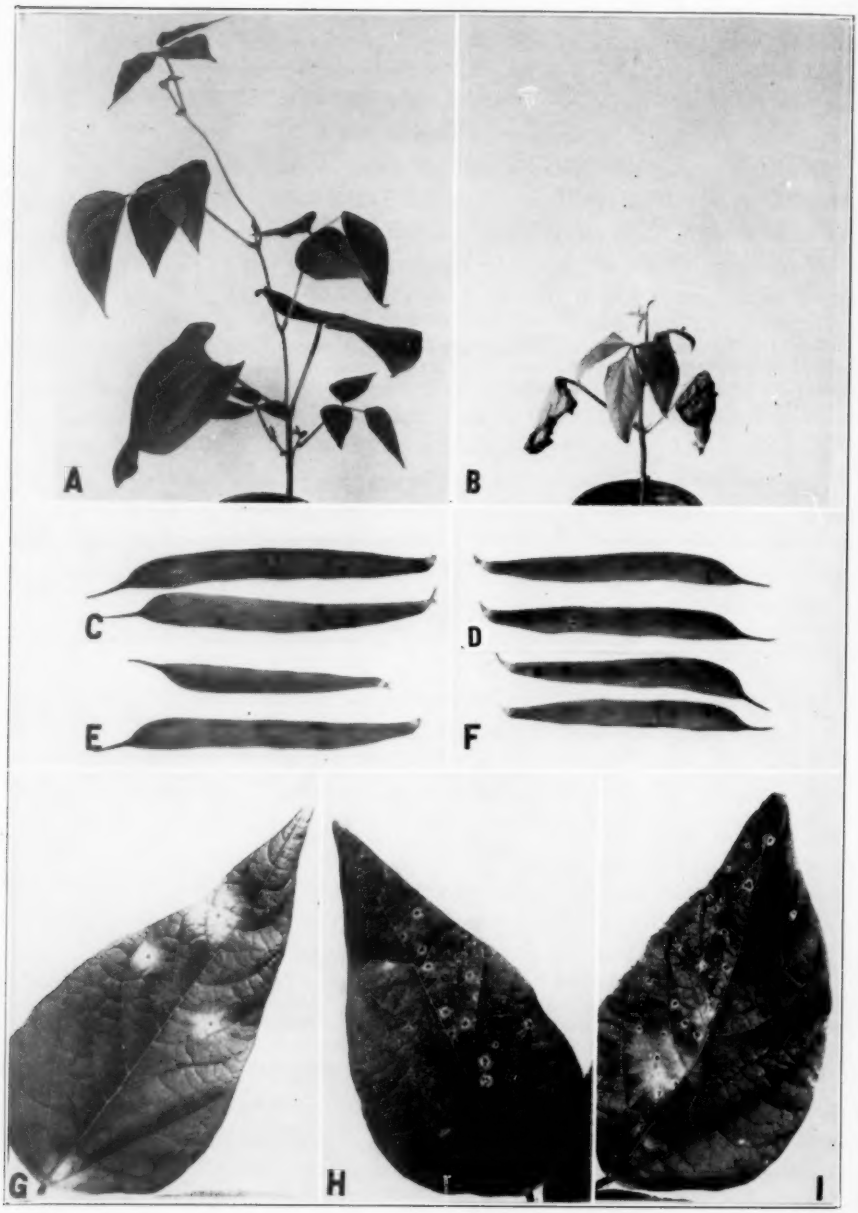

FIG. 1. Red Kidney plants, leaves and pods inoculated with various isolates of *Phytomonas medicaginis* var. *phaseolicola*. A. Eighteen days after stem inoculation with isolate 2079. Aside from a slight stunting and some faint mottling in the uppermost leaves, this plant showed no symptoms and grew as vigorously as uninoculated controls. B. Eighteen days after stem inoculation with isolate 2071. Severe stunting, wilting and vein-clearing symptoms are shown. C, D, E, and F. Pod lesions produced by spray inoculation with isolates 2076, 2079, 2074, and 2081, respectively. Pod lesions produced by isolates 2074 and 2079 were uniformly slightly smaller in size and those of 2079 were fewer in number than those produced by isolates 2076 and 2081. G, H, and I. Leaves spray-inoculated with isolates 2080, 2083, and 2077, respectively. The three leaves show (G) typical halo, (H) halo-less, and (I) both halo and halo-less lesions.

isolate producing only halo-less lesions regularly produced more lesions than any other isolate.

In some instances leaf inoculation of young, vigorously growing plants was followed by systemic infection. In all cases systemic infection was obtained only with isolates producing halo lesions. Thus the isolates varied in the number of lesions produced, in the time interval required to produce visible lesions, in the types of lesions produced, and in ability to produce systemic infection.

Stem Inoculations

Stem-puncture inoculations into young Red Kidney plants produced a wide range of symptoms. Inoculations made with a needle just below the primary leaves were usually carried out when the first trifoliate leaves were about one-half expanded. Usually each isolate was inoculated into 5 plants in each test.

Isolates producing halo or mixed type lesions on leaf inoculation, produced characteristic symptoms of the disease with stem inoculation. Four to 5 days after stem inoculation a slight wilting of the primary leaves occurred. By 6 days the first trifoliate leaflets, instead of being presented flatly, in a plane, more or less parallel to the ground line, were all pointing downward and exhibiting a slight flaccidity. This position is similar to that commonly assumed by bean leaflets at night. The reaction was especially noted in plants inoculated with some of the most virulent isolates. Six to 10 days after inoculation vein-clearing symptoms appeared. At about the same time wilting of one or more of the trifoliate leaves occurred. Within 15 to 20 days after inoculation the plants inoculated with virulent isolates were dead.

As a contrast to the extremely harmful effects just described, isolate 2075 under the same conditions produced no noticeable symptoms. Plants inoculated with this isolate could not be distinguished from healthy, uninoculated controls, except at the point of inoculation where slight discoloration and water-soaking of the nearby tissues occurred. Inoculated plants produced a set of pods which at maturity could not be distinguished from those on uninoculated controls.

Between these 2 extremes, other isolates produced some stunting, some wilting, and usually vein-clearing symptoms. However, the inoculated plants lived almost as long as control plants, which, although punctured with sterile needles, lived to complete maturity and produced a satisfactory crop of beans. In only one instance were vein-clearing symptoms produced by an isolate that produced only halo-less lesions on leaf inoculation.

Symptoms exhibited by plants inoculated by stem puncture thus fall into several groups. The first group consists of those showing marked stunting as early as 6 days after inoculation; and in these plants vein-clearing usually appeared at about the same time. A second group of isolates produced vein-clearing as early as 6 days after inoculation, but no noticeable stunting of inoculated plants occurred until several days later. In a third

group only extremely mild symptoms, such as a slight wilting of the primary leaves, or no symptoms at all, were produced. Photographs in figure 1, A, and 1, B, illustrate the range of symptoms obtained by stem inoculation on Red Kidney beans.

In some instances of stem inoculation a marked mottling appeared in the leaves near the growing point. This symptom occurred on fully grown plants that had been inoculated when young, but before any general yellowing or other similar signs of maturity appeared. Invariably the mottling symptoms occurred only with isolates that produced few or no typical symptoms of systemic infection.

Germinated Seed Inoculation

Seeds, germinated in rag-dolls at 28° C. for 66 hours, were placed in an aqueous bacterial suspension of each of the various isolates for a 4-hour period. At the end of this treatment each germinated seed was planted in soil in a porous clay pot. Usually, 10 germinated seeds were inoculated by the above method with each isolate.

All isolates produced typical water-soaked lesions on the cotyledons, and within 3 weeks every inoculated plant also showed halo-blight stem cankers. The isolates produced varying results in the development of systemic infection, as characterized by vein-clearing symptoms on the primary leaves and in the occurrence of premature death. Isolates that produced vein-clearing symptoms on the primary leaves produced death of the plants within the following 3-week period. Isolates that produced only halo-less lesions on leaf inoculation caused lesions on the cotyledons and cankers on stems with germinated seed inoculation, but did not produce vein-clearing symptoms of the primary leaves or any other symptom of systemic infection. In many instances the plants died before maturity, depending upon the severity of the stem cankers, but in other instances, in spite of stem cankers, the plants grew to maturity.

Pod Inoculations

A group of large, healthy Red Kidney plants with some of the pods on each plant nearly full-size and other pods still developing, was divided into 15 similar lots of 5 plants each. A water suspension of bacteria of each of the 13 isolates of the bacterium was sprayed on the pods of the respective lots with an atomizer. A check lot of 5 plants was sprayed with sterile water in each group of inoculations. After inoculation all plants were incubated in a moist chamber for 8 hours, after which they were placed on a greenhouse bench.

Lesions developed on pods of all lots within 4 to 8 days after inoculation. Final observations on pod infection were made 2 weeks after inoculation. Although leaf, stem, and germinated seed inoculations with the various isolates produced striking variations in symptoms, pod infection symptoms were all of the same general type, but varied in size and number of lesions. Several isolates, which were characterized by low pathogenicity in previous

comparative tests, produced lesions slightly smaller in diameter than those resulting from more virulent isolates. In general, a similar number of pods were infected, and approximately the same numbers of lesions were produced with each isolate although two isolates induced notably fewer lesions. None appeared on pods sprayed with sterile water.

Figures 1, C, D, E, and F, present photographs of pod lesions resulting from inoculation with several representative isolates.

EFFECT OF TEMPERATURE ON LESION TYPE

Because of the known effect (3) that high-temperature growing conditions have on the appearance of lesions of bean halo blight, it seemed desirable to study the effects of various temperatures on the lesions produced by several different isolates. Four isolates, 3 which produce only halo-less lesions and 1 which produces only typical halo lesions, were used in the study. Sixty young, healthy Red Kidney bean plants were divided into 4 similar lots of 15 plants each. Each lot was inoculated with a different isolate and then so distributed that the 5 plants inoculated with each isolate were held at 16, 22, and 28 degrees C. Table 2 records the results of this

TABLE 2.—*Effect of temperature on symptoms. Lesion type obtained when plant lots were leaf-inoculated with the indicated bacterial isolate and held at various temperatures*

Isolate number	Temperature °C.		
	16	22	28
2075	Halo-less	Halo-less	Halo-less
2079	Halo-less	Halo-less	Halo-less
2083	Halo-less	Halo-less	Halo-less
2076	Halo	Halo	Halo-less

test. It will be noted that the isolates producing halo-less lesions at 22° C. produced the same type of lesions at both 16° and 28°. In the case of the isolate producing typical halo lesions at ordinary temperatures, halo-less lesions resulted at 28° C. while at 16° and 22° typical halo lesions developed. This test confirms previous reports (3) to the effect that typical halo-blight isolates produce only halo-less lesions at 28° C., and it further shows that isolates causing halo-less lesions at ordinary temperatures (20–22° C.) also induce only halo-less lesions at 16 and 28°.

DIRECT INOCULATION FROM INFECTED TISSUE

In an attempt to determine whether or not bacteria from small, necrotic lesions would in turn produce only small, necrotic lesions, bacteria were transferred direct from small, necrotic or halo-less lesions to healthy, young plants. Other inoculations also were made with bacteria taken directly from typical halo lesions. Small areas of the leaf containing the lesions were cut out with sterile instruments and macerated in sterile mortars with sterile pestles. A water suspension of this material was then sprayed on leaves in

the manner described for leaf-inoculation technique. Necrotic lesions developed on leaves sprayed with bacterial suspension obtained from necrotic lesions. Halo lesions developed on leaves sprayed with a bacterial suspension of material from halo lesions. Occasionally, lesions, unlike those from which the inoculum was obtained, appeared in plants inoculated from either type of lesions. This would seem to indicate that variant strains of the halo-blight organism are continuously arising, or that mixtures were sometimes present in the lesions. Even in the latter case the presence of a mixture in the lesions would indicate that strain variations occur.

PHYSIOLOGICAL CULTURE STUDIES

The wide range of variation in symptoms obtained by leaf and stem inoculation of Red Kidney beans with the 13 halo-blight isolates suggested that such differences might be correlated with variations in physiological activity. Comparative studies on various types of media, commonly utilized for such observations, were carried out. In all experiments, observations were made on the 13 isolates of *Phytomonas medicaginis* var. *phaseolicola* together with an isolate of the bacterium causing common blight of beans, *P. phaseoli.*

The tests involved growth characteristics and rates-of-growth studies on beef extract agar streaks, beef extract broth, gelatin, plain milk, and milk with litmus and with brom-cresol-purple. Studies also were made on production of ammonia, hydrogen sulphide, starch digestion, and on the utilization of starch, dextrose, and sucrose. In all tests the various isolates of *Phytomonas medicaginis* var. *phaseolicola* showed characteristics that agreed with those described by Burkholder (1). In these tests the only variations obtained were slight differences in growth rates. These differences could not be correlated with those in pathogenicity.

DISCUSSION

The occurrence of a number of strains or variants of a given species or the variation in pathogenicity among isolates of a pathogenic species has been found the rule rather than the exception in biology. The discovery and description of such variation is now in itself of comparatively minor importance or significance. Frequently, however, a knowledge of the range of characteristics within which most members of a pathogenic species fall aids greatly in understanding more completely the organism and the disease produced by it. Such is the case in this report of the variation in symptoms produced on Red Kidney beans by various isolates of *Phytomonas medicaginis* var. *phaseolicola.*

With the discovery of physiological resistance in certain varieties to halo blight and the probable usefulness of such varieties in a breeding program, there has developed a need for a more thorough understanding of the various factors that influence the symptomatic picture of diseased plants in the field

and greenhouse. In a breeding program it is necessary to grow and inoculate large numbers of hybrid progenies. Selection of resistant plants must be made rapidly and usually without the benefit of laboratory pure-culture determinations of the causal organism. To do this, studies of the various factors responsible for symptom variations are essential.

This report now brings to three the number of factors shown to influence the symptoms of beans infected with halo blight. High temperatures were shown (3) to limit the formation of the typical halo development in susceptible varieties. Later, it was demonstrated (4) that certain varieties were physiologically resistant and manifested this resistance by the formation of small, necrotic, halo-less lesions instead of the large, characteristic halos commonly observed. This report now shows that certain isolates of the halo-blight organism may produce all halo or all non-halo or a mixture of these two types of primary lesions on susceptible varieties. In addition, the report describes systemic symptom differences that result from various isolates.

SUMMARY

Studies were made on the variations in symptoms produced on Red Kidney beans inoculated with 13 halo-blight (*Phytomonas medicaginis* var. *phaseolicola* Burkh.) isolates from naturally infected field-grown beans. Four inoculation techniques were used: leaf, stem, germinated seed, and pod. On the basis of pathogenicity exhibited in the various inoculation tests the isolates fall into 3 general groups. Four isolates were characterized by producing all halo or both halo and halo-less primary leaf lesions, typical pod lesions and marked stunting, wilting, vein-clearing symptoms and premature death with systemic infection. Another group of 4 isolates was characterized by the production of halo-less primary leaf lesions and small and reduced numbers of pod lesions. Stem inoculations with these same isolates produced little or no stunting or wilting and rarely vein-clearing symptoms or premature death. The third or intermediate group consisted of 5 isolates that usually produced mixed halo and halo-less primary leaf lesions and whose pathogenicity was intermediate between the two extreme groups just described.

In studies on the effect of temperature on primary lesions it was found that several isolates that induced only halo-less primary lesions at 22° C. also induced only halo-less lesions at 16° and 28°, as contrasted with the behavior of other isolates, which produced halo-less lesions at 28° but produced typical halo lesions at 16° and 22° C.

Water suspensions of macerated tissue from typical halo lesions when sprayed on young leaves caused typical halo lesions, whereas bacteria from halo-less lesions caused halo-less lesions. Occasionally, lesions unlike those from which the inoculum was obtained appeared in the inoculated plants.

In all physiological culture tests the various isolates showed characteristics that agreed with those described by Burkholder (1). In these tests the

only variations obtained were slight differences in growth rates, which could not be correlated with differences in pathogenicity.

DEPARTMENT OF PLANT PATHOLOGY,
NEBRASKA AGRICULTURAL EXPERIMENT STATION,
LINCOLN, NEBRASKA.

LITERATURE CITED

1. BURKHOLDER, W. H. The bacterial diseases of the bean. Cornell Univ. Agr. Exp. Stat. Memoir 127. April, 1930.
2. Committee on Bacteriological Technique of the Society of American Bacteriologists. Pure Culture Study of Bacteria. Vol. VII, No. 4. Leaflet V, 7th ed., 1931.
3. GOSS, R. W. The relation of temperature to common and halo blight of beans. Phytopath. **30**: 258–264. 1940.
4. JENSEN, J. H., and R. W. GOSS. Physiological resistance to halo blight in beans. Phytopath. **32**: 246–253. 1942.
5. SHERF, A. F. A method for maintaining *Phytomonas sepedonica* in culture for long periods without transfer. Phytopath. **33**: 330–332. 1943.

SEEDLING INVASION OF THE COVERED SMUT OF OATS

PAUL F. BRANDWEIN

(Accepted for publication December 21, 1943)

The behavior and sexuality of the various oat smuts in culture have been subjected to considerable investigation. Many authors, Dickinson (4), Holton (5, 6), and Utter (12), have emphasized the heterosexuality of the oat smuts, and there is considerable evidence to show that monosporidial lines do not infect the host, but that fusion of the sporidia of presumably different sexes result in an infection hypha. Western (11) studied, in *Ustilago avenae*, the behavior of 2 monosporidial lines of unlike sex introduced by pressure under the glumes of oat seeds. Under these more natural conditions, there was a union of adjacent promycelial segments by means of a fusion tube, which finally gave rise to the true infection hypha. Tisdale and Tapke (8), Tapke (9), Kolk (7), and Western (10) also have described the invasion of different hosts by the promycelium directly produced from chlamydospores. Since 1936, the results of experiments designed to discover to what extent seedlings are invaded by the promycelia from the chlamydospore of the covered smut of oats have been carried on and are reported here.

EXPERIMENTAL

The covered smut *Ustilago levis* (*U. Kolleri*), race 7, and the oats, var. Monarch, which is 100 per cent susceptible, and Markton, which is completely resistant to it, were used. Kolk (7), Western (10) and Brandwein (1, 2), have proved beyond doubt that the smut penetrates resistant and susceptible coleoptiles alike. In Markton the inoculation results definitely in seedling invasion and sometimes in non-sporulating infection (Brandwein (3)).

Dehulled seeds of Monarch and Markton were dusted with dry chlamydospores and the seeds were then germinated in sand having a moisture content of 20 per cent of its water-holding capacity. This sand was contained in paper cups and maintained at 20° C. Twenty-four-hour-old seedlings also were thoroughly dusted with spores, and new sand of the proper moisture content was then replaced. The plants inoculated at the seed stage were removed after 48 hours for examination, the others inoculated at 24 hours were removed at 72 hours. Most useful preparations were made by stripping the coleoptile with forceps and examining the unstained tissue by suitable lighting under 440 and 950 (oil immersion) magnification. Successive strips from one plant permitted a fairly thorough examination of from $\frac{1}{3}$ to $\frac{1}{2}$ of the outer coleoptile tissue. The results are tabulated in table 1.

It was surprising to find that of the tremendous numbers of spores found on a seedling inoculated at 24 hours there were very few germinations—roughly 10 per cent—and few cases of what could be definitely called invasion.

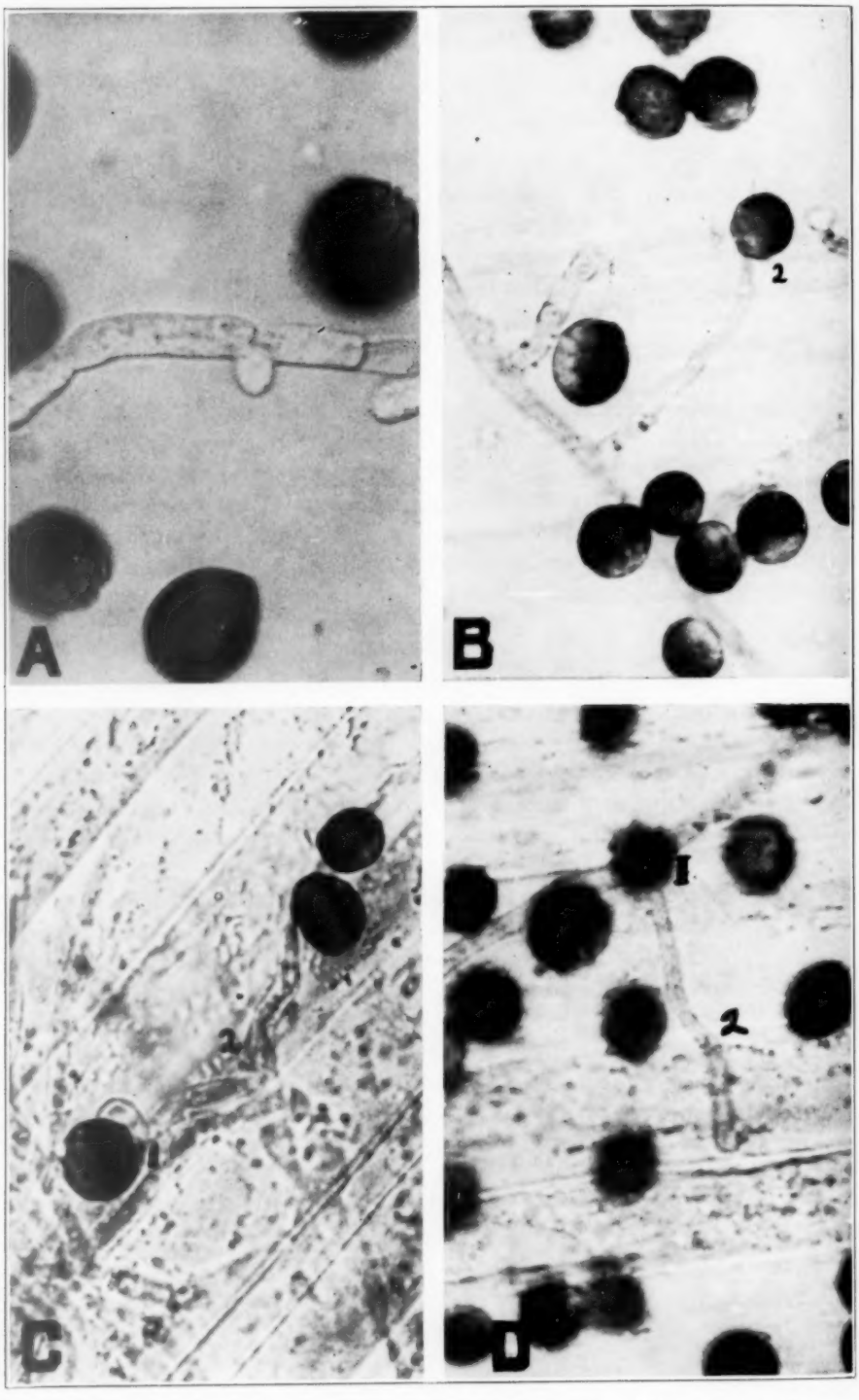

FIG. 1. Chlamydospore germination and promycelia invasion of the oat coleoptile.

Fusion of sporidia was relatively uncommon. Of the 532 cases of hyphal penetration that seemed to result from fusion, only 52 seemed to be the result of fusion of sporidia or sporidia-like structures.

Formation of sporidia (Fig. 1, A) and sporidial fusions were, however, commonly found in the mat of smut spores that sometimes was found in the sand alongside the seed.

Figure 1, B, shows a type of penetration (in Monarch) commonly found; the chlamydospore promycelium enters at 2, produces a ramifying mycelium internally which finally dips into the tissues. On the other hand, Fig. 1, C, shows 2 penetrations in Monarch from 2 promycelia, with the external and

TABLE 1.—*Spore-germination and host-invasion results obtained from oat seedlings grown from dehulled seeds of Markton and Monarch oats that had been dusted with chlamydospores of Ustilago levis*

No. seedlings examined	Host	Age of seedling (hours)	No. of invasions	Invasions directly from chlamydospore	Doubtful[a] or from fused hyphae
47	Monarch	48	201	71	130
46	"	72	407	219	188
23	Markton	48	130	32	98
27	"	72	184	68	116
143			922	390	532

[a] In 286 cases, it could not be readily determined, because of ramification and branching of the mycelium, whether penetration was that of a hypha formed by fusion of elongated promycelial segments or that of the promycelium.

internal mycelia ramifying and bunching in and on the coleoptile. In this type it was difficult to determine whether the penetration was directly from a chlamydospore or from fusion at 1 and 2 (Fig. 1, C). Fig. 1, D, shows the internal mycelium from the spore 1 in a Markton coleoptile. It is interesting to note a swelling of the cell wall just in front of the penetration hypha, which had entered at 2. Western (11) has described similar pads of cellulose. The writer has observed 3 more cases similar to this in living material. It is obvious that such favorable material would be difficult to obtain. Figures 1, B, and 1, C, are typical of the rich mycelium that quickly develops in susceptible plants; and Fig. 1, D, is typical of the sparse mycelium within the coleoptile cells of the resistant plants.

SUMMARY AND CONCLUSION

In this paper, the writer reports and figures extensive penetration by the promycelium arising from the chlamydospore of *Ustilago levis*. No mycelial fusions were apparent in these cases. Does the dikaryophase arise from such a direct penetration? At present no evidence is available. There is also no present way of knowing that these promycelia produce sporulating infection. But it is true that these direct penetrations appear to be more numerous than might be expected.

The problem deserves further study. The technique of examining invasion hyphae needs refinement, and the progress of invasion hyphae should be studied by film, if possible.

In conclusion, it may be stated that under the environmental conditions reported here, penetration by the promycelium directly from the chlamydospore of *Ustilago levis*, race 7, without promycelial fusions, is more extensive than has been supposed. It is suggested that the story of the development of the oat smut chlamydospore on the coleoptile may be different in certain important respects from its development in culture.

Thanks are due to Dr. G. M. Reed for the materials with which this work has been carried on and for the advice he has given so generously.

BROOKLYN BOTANIC GARDEN,
BROOKLYN, N. Y.

LITERATURE CITED

1. BRANDWEIN, P. F. Experiments on latent infection of resistant varieties by the loose and covered smuts of oats. Bull. Torr. Bot. Club **64**: 433–444. 1937.
2. ———. The emergence of smut-inoculated oat seedlings through sand and loam soil. Bull. Torr. Bot. Club **65**: 477–483. 1938.
3. ———. Infection studies on the covered smut of oats. Bull. Torr. Bot. Club **67**: 673–689. 1940.
4. DICKINSON, S. Experiments on the physiology and genetics of smut fungi. Seedling infection. Proc. Roy. Soc. (London) **102**: 174–176. 1927.
5. HOLTON, C. S. Hybridization and segregation in the oat smuts. Phytopath. **21**: 835–842. 1931.
6. ———. Further studies on the oat smuts, with special reference to hybridization, cytology, and sexuality. Jour. Agr. Res. [U. S.] **62**: 229–240. 1941.
7. KOLK, M. Relation of host and pathogen in the oat smut, *Ustilago avenae*. Bull. Torr. Bot. Club **57**: 443–507. 1930.
8. TISDALE, W. H., and V. F. TAPKE. Infection of barley by *Ustilago nuda* through seed inoculation. Jour. Agr. Res. [U. S.] **29**: 263–284. 1924.
9. TAPKE, V. F. Occurrence, identification, and species validity of the barley loose smuts, *Ustilago nuda*, *U. nigra*, and *U. medians*. Phytopath. **33**: 194–209. 1943.
10. WESTERN, J. H. The biology of the oat smuts, IV. The invasion of some susceptible and resistant oat varieties, including Markton, by selected biological species of smut, *Ustilago avenae* (Pers.) Jens. and *Ustilago Kolleri*, Wille. Ann. App. Biol. **23**: 245–263. 1936.
11. ———. Sexual fusion in *Ustilago avenae* under natural conditions. Phytopath. **27**: 547–553. 1937.
12. UTTER, L. G. Culture and inoculation studies on races of the loose and covered smut of oats. Amer. Jour. Bot. **25**: 198–209. 1938.

PATHOGENESIS OF APHANOMYCES COCHLIOIDES ON TAPROOTS OF THE SUGAR BEET[1]

W. F. Buchholtz and C. H. Meredith[2]

(Accepted for publication January 3, 1944)

Since the discovery that *Pythium debaryanum* Hesse is the principal cause of damping-off of very young sugar-beet seedlings in northern Iowa, the major role played by another pathogen in parasitizing older seedlings and plants with taproots of considerable size, in addition to young seedlings, under certain conditions in specific entire fields, has become increasingly evident.[3] Infection of plants at and beyond the seedling stage occurred in two adjacent fields on the Northern Iowa Experimental Association Farm (Fig. 1) in 1937 and in 1938. Isolation and inoculation trials indicated that *Aphanomyces cochlioides* Drechsler was the causal agent. Since some of the symptoms observed in the field and those induced by pure culture inoculations were on plants beyond the seedling stage, and, therefore, in addition to those hitherto attributed to *A. cochlioides*, they are herein briefly described.

SYMPTOMS

On young seedlings the symptoms were similar to those attributed to *Aphanomyces cochlioides* by Peters,[4] Edson,[5] and Drechsler.[6] Occasionally a general rapid necrosis of the entire root, hypocotyl, and the lower portion of the cotyledons of young seedlings occurred (Fig. 2, A). *A. cochlioides* apparently parasitizes very young seedlings less frequently than *Pythium debaryanum*. Furthermore, under field conditions, direct necrosis induced by *P. debaryanum* is restricted to that portion of seedlings below the soil surface, with brown rather than black discoloration of necrotic tissues.

On older seedlings or plants with 1 or 2 other leaves and with some secondary vascular enlargement there was necrosis of the cortex of roots and hypocotyls and of the cotyledonary petioles (Fig. 2, B). The necrotic tissues were black; in fact, this is the typical so-called "black-root" in northern Iowa. In this late seedling stage of development, the surfaces of hypocotyls and primary roots in *Aphanomyces cochlioides*-infested soil were rough and gray.

A later and very striking symptom induced by *Aphanomyces cochlioides* was a black disintegration of the taproot, which first became manifest in late

[1] Journal Paper No. J–1175 of the Iowa Agricultural Experiment Station, Ames, Iowa. Project No. 432.

[2] The writers are indebted to Dr. I. E. Melhus for aid and suggestions during the course of investigations and preparation of the manuscript.

[3] Buchholtz, W. F., and C. H. Meredith. A sugar-beet root rot caused by *Aphanomyces cochlioides*. (Abst.) Phytopath. 28: 4. 1938.

[4] Peters, L. Zur Kenntnis des Wurzelbrandes der Zuckerrübe. Ber. Deut. Bot. Gesell. 24: 323–329. 1906.

[5] Edson, H. A. Seedling diseases of sugar beets and their relation to root-rot and crown-rot. Jour. Agr. Res. [U. S.] 4: 135–168. 1915.

[6] Drechsler, Charles. The beet water mold and several related root parasites. Jour. Agr. Res. [U. S.] 38: 309–361. 1929.

June and early July, when the thinned stands were recovering from the thinning operation (Fig. 2, C). The pathologic events leading to this condition are not entirely understood. There is likelihood of earlier infection and pathogenesis, for crowns of afflicted plants were considerably stunted and their lower leaves yellow in early stages of indirect necrosis. The central leaves were sometimes unusually stiff and glossy but dwarfed. Wilting in the afternoon of bright days, with subsequent recovery ("flagging") was common. The roots of such plants were undersized, black-tipped 3 to 6 inches below the soil line, with an excess of side roots developed apparently

FIG. 1. A field of sugar beets which was a complete failure because of "tip rot" caused by *Aphanomyces cochlioides*. Kanawha, Iowa, 1937. (Photograph by I. E. Melhus.)

in response to the destruction of the taproot (Fig. 2, C). A large majority of these side roots were black and shrivelled.

After mid-June, plants in infested soil either were killed or grew very slowly until late August, when there was some recovery and apparently satisfactory growth of many of the remaining plants. Even then, some plants underwent a progressive rot of the fleshy taproot at about the plow line. This rot was characterized by a first greenish-yellow, later light-brown and finally dark-brown, almost black, discoloration (Fig. 2, D). Infected tissues were slightly softened but still resisted the knife, and split rather than tore when cut. They appeared slightly water-soaked. When subject to desiccation such tissues finally shrivelled to a "tassel" of vascular elements (Fig. 2, E and F) as in the rot produced by *Phytophthora drechsleri*

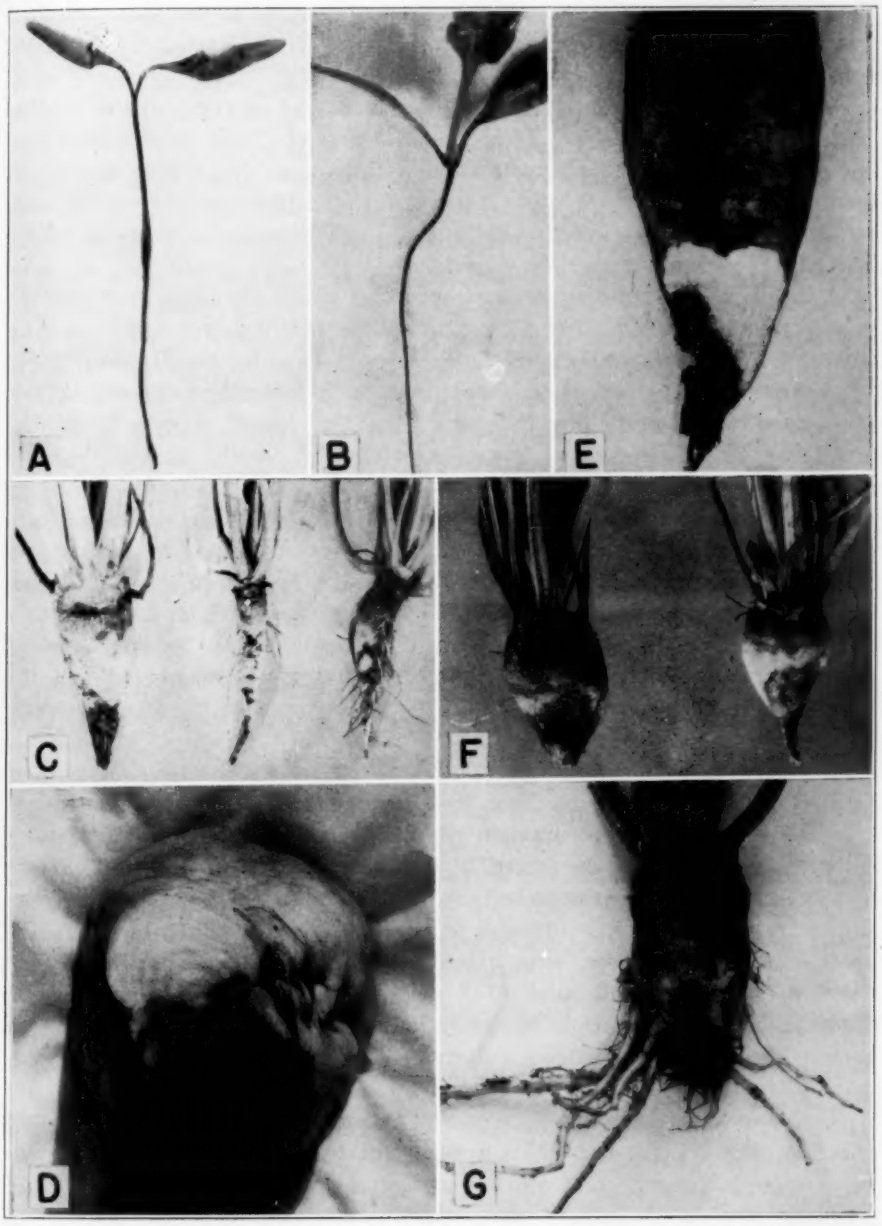

FIG. 2. A. Seedling that has undergone necrosis of the entire root, hypocotyl and lower portion of the cotyledons. B. Seedling with darkened hypocotyl and cotyledonary petioles ("black root"). This symptom is a reliable diagnostic characteristic of *Aphanomyces cochlioides* infection of beet seedlings. C. Tip-rotted beets about a month after thinning. The blackened taproots are typical as is the excessive development of side roots (also blackened) on the beet at the right. D. Typical watersoaked but firm, discolored area induced by *A. cochlioides* on large taproots. This root was exposed to an agar culture of *A. cochlioides*. E. Section cut through rotted, partially desiccated lower taproot. F. "Tassels" left on the taproot when tissues invaded by *A. cochlioides* become desiccated. G. An apparently "recovered" beet from a field that was a failure because of tip rot. Note the several horizontal roots rather than a taproot, and the blackened small roots.

Tucker.[7] Frequently the uppermost progress of rot of the fleshy taproot was in the two zones of lateral root protrusion. There was no distinct odor associated with rotted fleshy taproots.

Late in the season many remaining beets showed signs of previous infection and partial recovery. After the destruction of the original taproot, one or several of the side roots appeared to have achieved some degree of dominance and developed into a taproot, frequently in a more nearly horizontal than a vertical direction (Fig. 2, G). The vascular elements of the original taproot sometimes were in evidence.

Because the destruction of the taproot at about the plow line was the most common and characteristic symptom on plants beyond the seedling stage, "tip rot" became the common field designation for the disease.

Aphanomyces cochlioides appeared more or less regularly from plantings of diseased tissues on plain agar from the first time isolations were attempted in August, 1937. On August 21, two growths of *A. cochlioides* were detected in a series of plantings of dry, blackened side roots on plain agar. On August 23, *A. cochlioides* was detected in 6 of a series of isolations from rotten taproots. On August 31, 11 of 12 isolations from wet rotted taproots yielded *A. cochlioides*, and on September 21, 23 of 24 such isolations did also. Since then the same pathogen has been observed repeatedly and consistently in isolations from seedlings and plants of all sizes with the various types of symptoms described. Except for slightly smaller vegetative hyphae, the isolates obtained seemed to fall within the limits described by Drechsler.[8]

INOCULATION TRIALS

In 3 inoculation experiments in 1937, *Aphanomyces cochlioides* produced typical taproot-rot symptoms. In the first trial, September 11, corn-meal cultures of each of 2 isolates were introduced by means of plug wounds into 5 beets in the field. In the second, on September 20, each of 6 isolates were similarly introduced into 10 beets in the field. On October 14, when the beets in both experiments were lifted from the soil, all inoculated beets in the first experiment and 55 of 60 in the second were partially rotted. On September 29, each of 8 isolates was introduced by plug wounds into 3 lifted beets. The inoculated beets were stored in the greenhouse until November 1, when 23 had rotted. In all 3 experiments, the checks were not rotted, and *A. cochlioides* was recovered from the inoculated rotted beets. On two other occasions, sugar-beet roots were inoculated with isolates of *A. cochlioides* in the laboratory, and necrosis of tissues developed.

Four inoculation experiments were undertaken in 1938. Two were started in an April-planted field on July 12 and August 5. Ten plugged and 5 unwounded beets were exposed to each of 6 isolates of *Aphanomyces cochlioides*. Two similar unexposed series served as checks. The beets were lifted and examined on August 19. From the July 12 inoculations, only 2

[7] Tompkins, C. M., B. L. Richards, C. M. Tucker, and M. W. Gardner. Phytophthora rot of sugar beet. Jour. Agr. Res. [U. S.] 52: 205–216. 1936.

[8] See footnote 6.

of 60 plugged beets showed typical rotting; none of the uninjured beets showed any evidence of infection. From the August 5 inoculations, 2 isolates typically rotted all 10 plugged beets, a third isolate rotted 5 of 10 plugged beets, a fourth isolate rotted 1 of 10 plugged beets, the other 2 isolates rotted none. One of the 30 unwounded, exposed beets showed evidence of typical rotting by *A. cochlioides*. One of the beets that rotted as a result of exposure to a virulent isolate is shown in figure 2, D.

In a third similar experiment, started August 19, only the 4 apparently most virulent isolates were used. One isolate, which rotted an unwounded beet in the August 5 inoculation, typically rotted all 10 plugged beets; the 3 other isolates rotted 8, 7, and 4 plugged beets, respectively. No unwounded exposed beets were rotted. All checks remained healthy.

A fourth experiment was begun on August 18. Three sets of 4 injured (3 scratches with knife blade) and 4 uninjured beets of 4 ages (planted March 29, April 12, May 15, June 4) were exposed to 2 virulent isolates of *Aphanomyces cochlioides*. On September 9 the beets were pulled and the following observations made: Five of 16 injured, exposed beets from the two first plantings were slightly rotted on the surface only. Four of 8 injured beets in the third planting were similarly surface-rotted; one other had typical "tip rot." In the first 3 plantings, no uninjured exposed beets showed any evidence of infection as indicated by rotting. Of the youngest beets (June 4 planting) 7 injured and 7 uninjured roots showed some rot. Obviously roots of the last planted beets were rotted more after exposure to *A. cochlioides* than those of the larger beets in the first 3 plantings. All roots exposed to sterile agar were healthy.

In several trials, isolates of *Aphanomyces cochlioides*, when added to steamed soil, caused damping-off of beet seedlings growing in such soil. The invaded necrotic seedling tissues invariably were black, and the cotyledonary petioles were typically invaded (Fig. 2, A). In each field inoculation trial in 1938, and in two soil infestation trials in the greenhouse, *A. cochlioides* was recovered from one or more rotted roots or damped-off seedlings.

SUMMARY

The pathogenic effects of *Aphanomyces cochlioides* on sugar beet plants at and beyond the young seedling stage were observed in northern Iowa and are recorded. The symptoms were necrosis and blackening of hypocotyls, cotyledonary petioles and roots of seedlings; blackening and disintegration of the taproot 3 to 6 inches below the soil line at thinning time, with wilting and indirect necrosis of leaves; necrosis and blackening of excessive side roots; a greenish-yellow to brown or black semi-soft rot of the lower end of large taproots. *Aphanomyces cochlioides* was isolated frequently from such tissues.

Inoculation experiments with pure cultures induced symptoms similar to those observed in the field. In one inoculation trial with beets of 4 ages, young roots seemed to be more susceptible than older roots.

THE SEQUENCE OF INFECTION OF A SEEDLING STAND OF SUGAR BEETS BY PYTHIUM DEBARYANUM AND APHANOMYCES COCHLIOIDES[1, 2]

W. F. Buchholtz

(Accepted for publication January 13, 1944)

The role of *Pythium debaryanum* Hesse in destroying germinating seed and young seedlings of sugar beets in northern Iowa is well known.[3] Seed treatment also is recognized as valuable and is practiced universally among sugar beet growers. Although seed treatment has been effective in establishing good stand of young seedlings, these stands often have failed soon after their establishment. It is known that *Aphanomyces cochlioides* Drechsler can attack the roots of older seedlings and of larger plants.[4]

In this presentation an attempt is made to delineate the roles of *Pythium debaryanum* and *Aphanomyces cochlioides* in destroying seedling stands in northern Iowa soils and to evaluate seed treatment as a control for each.

METHODS

In September, 1938, samples of Clarion loam and Webster loam soils were taken from a field that had been in alfalfa for 3 years and that had not grown beets since 1932. Beets growing in this field in 1939 showed no evidence of infection by *Aphanomyces cochlioides*, although the seedling stand loss from *Pythium debaryanum* was heavy on the Clarion loam and light on the Webster loam. A comparable sample of Webster loam was taken from an adjacent field in which beets were heavily diseased at all ages by *A. cochlioides* and considerable numbers of young seedlings destroyed by *P. debaryanum*. The pH values of the 3 samples were 5.5, 6.6, and 6.3, respectively. The samples were stored in the greenhouse as taken from the field in galvanized tin cans, covered but not sealed. Normal greenhouse temperatures prevailed, and the samples did not become dry.

Five 4-inch pots were filled from each of the 3 different soil samples, in January, 1939. A row of 10 nontreated and a row of 10 treated (New Improved Ceresan at the rate of 5 oz. per 100 lb. of seed) clusters of American No. 1 seed were planted in each pot. In addition to the above samples, 5 pots of the Webster loam infested with *Pythium debaryanum* and *Aphanomyces cochlioides* were steamed and likewise planted. Germination and number of diseased seedlings in each pot were recorded daily for 28 days. Diseased seedlings were removed every day, washed, and laid on

[1] Journal Paper No. J–1171 of the Iowa Agricultural Experiment Station, Ames, Iowa. Project No. 432.

[2] The writer is indebted to Dr. I. E. Melhus for helpful suggestions in connection with the investigation and in the preparation of the manuscript.

[3] Buchholtz, W. F. Factors influencing the pathogenicity of *Pythium debaryanum* on sugar beet seedlings. Phytopath. 28: 448–475. 1938.

[4] Buchholtz, W. F., and C. H. Meredith. Pathogenesis of *Aphanomyces cochlioides* on taproots of sugar beet. Phytopath. 34: 485–489. 1944.

plain agar. The pathogens grew out in 24 to 48 hours and were identified and recorded. Of 245 seedlings becoming diseased, diagnosis was successful with all but 4. *P. debaryanum* produced sphaerosporangia and occasional oogonia, antheridia, and oospores in 48 hours. *A. cochlioides* exhibited typical coarse, tortuous mycelium and occasionally the first stage of zoospore formation in 8 to 24 hours.

EXPERIMENTAL RESULTS

In steamed soil the rate and extent of germination of treated and nontreated seed were about the same. Germination was nearly complete 11

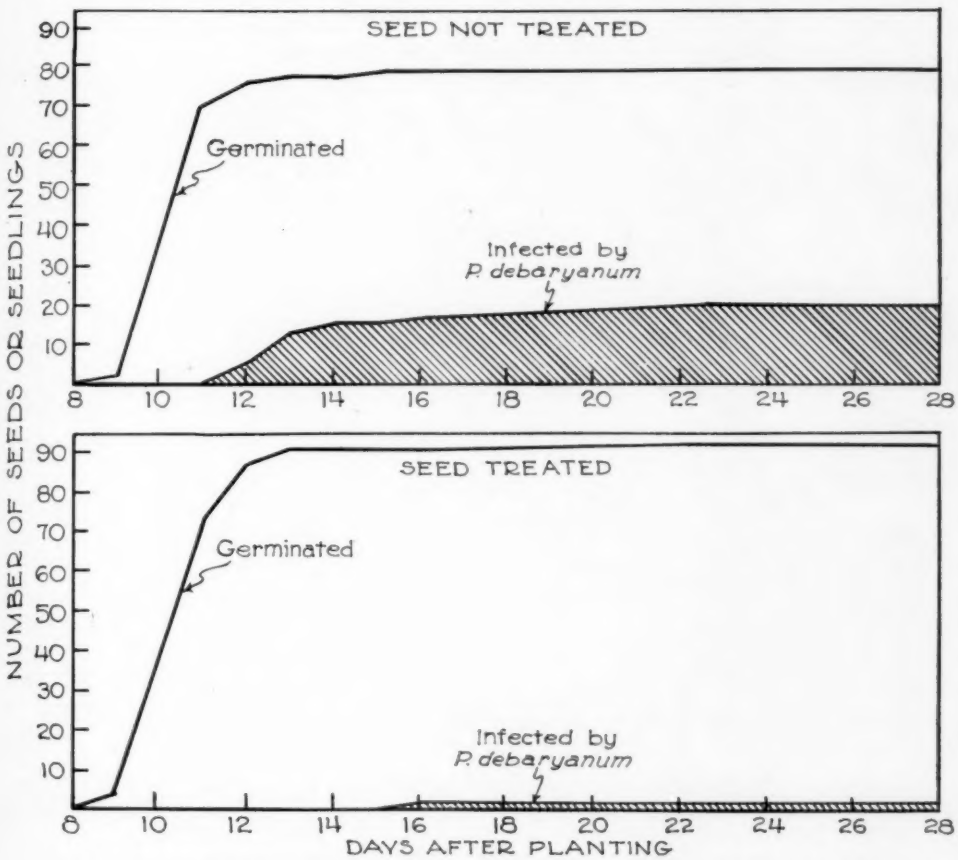

FIG. 1. The number of seeds germinated and seedlings infected daily from 50 nontreated and 50 treated seed clusters planted in the greenhouse in Webster loam lightly infested with *Pythium debaryanum*.

days after planting, and the final numbers of seedlings from 50 nontreated and 50 treated seed clusters were 90 and 86, respectively. No seedlings were diseased (Fig. 4, A). This lot of seed apparently was free from *Phoma betae*.

In soil lightly infested with *Pythium debaryanum* (Webster loam, alfalfa sod), germination was nearly completed 12 days after planting (Fig. 1).

Germination was less from nontreated than from treated seed, which probably was due to interruption of the germination of nontreated seed by *P. debaryanum*. Loss of seedlings from infection by *P. debaryanum* occurred soon after germination, and there was little or no loss beyond the 14th day after planting, when there were 63 and 91 healthy seedlings remaining from nontreated and treated seed, respectively (Fig. 4, B). Twenty seedlings grown from the nontreated seed were killed. Since ap-

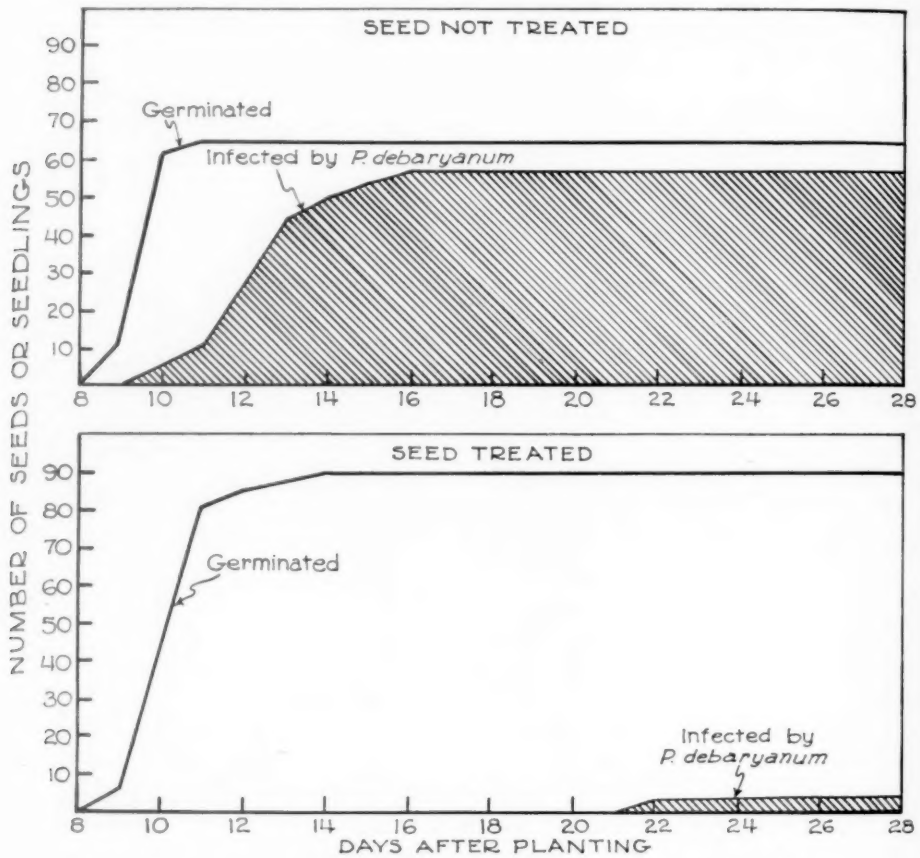

FIG. 2. The number of seeds germinated and seedlings infected daily from 50 nontreated and 50 treated seed clusters planted in the greenhouse in Clarion loam heavily infested with *Pythium debaryanum*.

proximately 10 germinating seeds probably were destroyed, the loss was $\frac{1}{3}$ the expected stand of approximately 90. Only 1 seedling from treated seed was destroyed, also by *P. debaryanum*.

In soil heavily infested with *Pythium debaryanum* (Clarion loam, alfalfa sod), germination was nearly completed 11 days after planting (Fig. 2). A total of 65 seedlings resulted from nontreated seed and 90 from treated seed, a difference of 25 as compared with a difference of 13 in the Webster loam, lightly infested with *P. debaryanum*. Here again the loss of seed-

lings grown from nontreated seed occurred soon after germination. Most of the loss had taken place by the 14th day and none beyond the 16th day after planting, when there were 7 and 90 healthy seedlings remaining from nontreated and treated seed, respectively (Fig. 4, C). Fifty-seven seedlings from nontreated seed were infected by *P. debaryanum*. Since 25 germinating seeds probably were destroyed, there was a total loss of 82 seeds and seedlings, or fully 90 per cent of the probable number of live seeds. Only 4 seedlings from treated seed were infected by *P. debaryanum*.

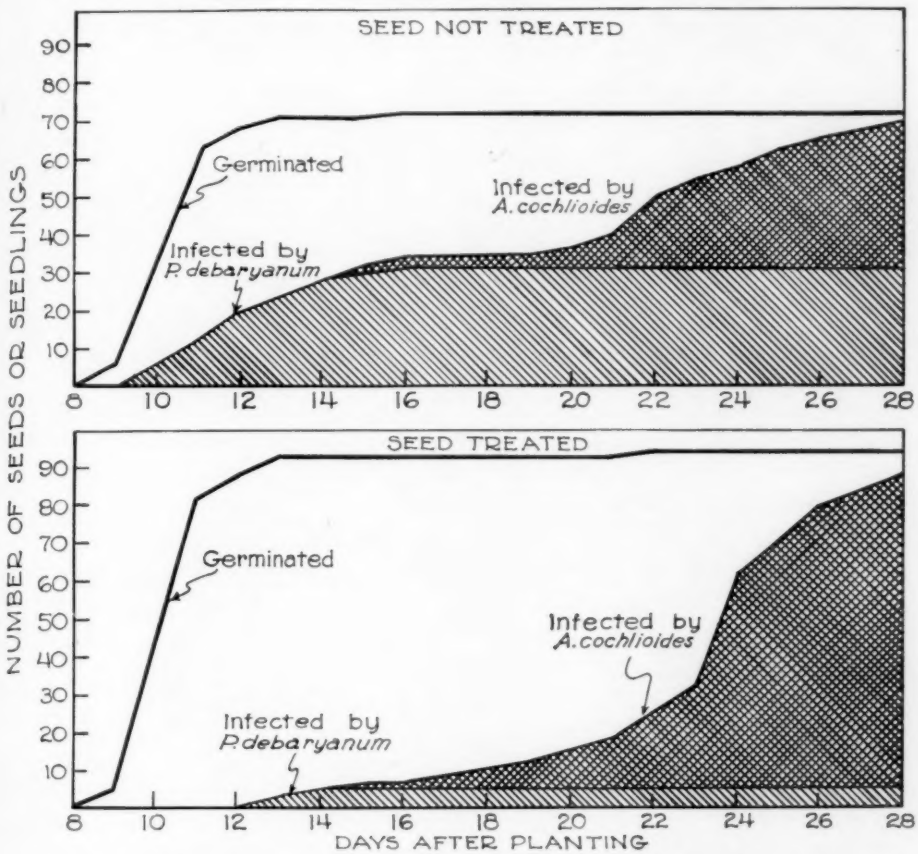

FIG. 3. The number of seeds germinated and seedlings infected daily from 50 treated and 50 nontreated seed clusters planted in the greenhouse in Webster loam heavily infested with *Pythium debaryanum* and *Aphanomyces cochlioides*.

In soil infested with *Pythium debaryanum* and *Aphanomyces cochlioides* (Webster loam), germination was nearly completed 12 days after planting (Fig. 3). There were, in all, 72 seedlings from nontreated seed and 94 from treated seed, a difference of 22 as compared with 13 in the lightly infested Webster loam and 25 in the heavily infested Clarion loam. Loss of seedlings from nontreated seed by *P. debaryanum* infection again was rapid. Nearly all infection had occurred by the 15th day after planting, and none occurred

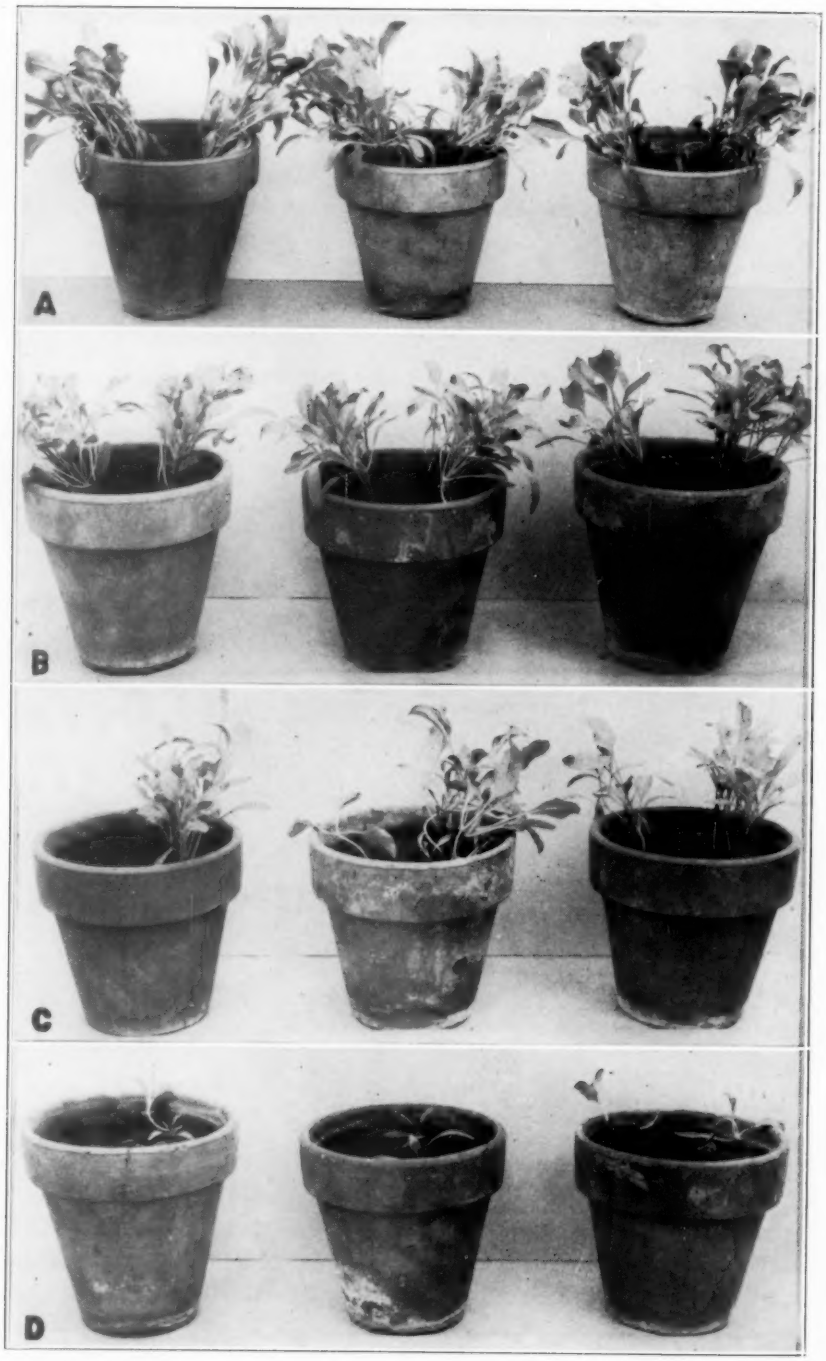

FIG. 4. Typical final stands of sugar beet seedlings grown in (A) steamed Webster loam, (B) Webster loam lightly infested with *Pythium debaryanum*, (C) Clarion loam heavily infested with *P. debaryanum*, and (D) Webster loam infested by *P. debaryanum* and *Aphanomyces cochlioides*. In each pot the left row is from nontreated seed, the right row from treated seed.

after the 16th day. Thirty-one seedlings from nontreated seed were infected by *P. debaryanum* in addition to the 22 germinating seeds probably destroyed by this pathogen. Thus, a total of 53 seeds and seedlings, or nearly 60 per cent, probably were killed by *P. debaryanum*. The interruption in germination in this case was assumed to have been caused by *P. debaryanum*, since the loss of seedlings from infection by *P. debaryanum* had ceased by the time the first seedlings were visibly infected with *Aphanomyces cochlioides*. Only 5 seedlings from treated seed were infected by *P. debaryanum*, all by the 15th day after planting.

In stands from treated and nontreated seed loss of seedlings from infection by *Aphanomyces cochlioides* began on the 15th day after planting and continued until the 28th day after planting, when there remained only 1 healthy seedling from nontreated seed and 3 from treated seed (Fig. 4, D).

FIELD OBSERVATIONS

In the spring of 1939, plantings of treated and nontreated seed were made in the field on the areas from which the soil samples had been taken for the greenhouse tests. Isolation from every infected seedling, as in the greenhouse test, was impossible because many died and dried up before they were picked up, even though the stands were observed daily. Isolations were made daily, however, from typically diseased seedlings from treated and nontreated seed in the 3 areas. In general, as in the greenhouse tests, stands from nontreated seed in the heavily infested Clarion loam and in the lightly infested Webster loam were reduced more by *Pythium debaryanum* than were the stands from treated seed in the same areas. In the soil infested with *Aphanomyces cochlioides*, however, nearly the entire stand, whether from treated or nontreated seed, became infected by this pathogen.

The general infection of beet stands in commercial fields infested with *Aphanomyces cochlioides* is a common occurrence. In many fields stands of young seedlings may be excellent, which, in some cases, doubtless is the result of protection from *Pythium debaryanum* by seed treatment. It has been repeatedly observed, however, that in field stands, as in the greenhouse cultures, *A. cochlioides* destroys plants that have developed beyond the very young seedling stage, at a time when a seed disinfectant and protectant cannot be expected to persist and protect the growing plant.

SUMMARY

Pythium debaryanum killed 33 per cent of the sugar beet seedlings grown from nontreated seed in lightly infested soil, 90 per cent in heavily infested soil, and 60 per cent in soil in which *P. debaryanum* and *Aphanomyces cochlioides* occurred together. The killing of seedlings by *P. debaryanum* was nearly completed 15 days after planting. Very few of the seedlings that developed from treated seed were killed by *P. debaryanum*.

Aphanomyces cochlioides infection began when the seedlings ceased

dying from *Pythium debaryanum* infection, about 15 days after planting. By the 28th day after planting nearly all the remaining live seedlings, whether from treated or nontreated seed, had been attacked by *A. cochlioides.*

Pythium debaryanum took its toll quickly and was readily controlled by seed treatment, whereas *Aphanomyces cochlioides* attacked older seedlings and was not controlled by seed treatment.

AGRICULTURAL EXPERIMENT STATION,
BROOKINGS, SOUTH DAKOTA.

ORNITHOGALUM MOSAIC

FLOYD F. SMITH[1] AND PHILIP BRIERLEY[2]

(Accepted for publication January 12, 1944)

A double-flowered variety of *Ornithogalum thyrsoides* Jacq. (Liliaceae) was received from a grower in Oregon in 1940. All plants from the original sample of bulbils exhibited mild green mottling in leaves and prominent light- and dark-green blotching in flower stalks. Seedlings of the same species grown from commercial seed were single-flowered and free from mottling. The disease in the Oregon specimens was investigated to determine its relation to the virus diseases of lily, tulip, hyacinth, and onion.

The only previous record of mosaic in *Ornithogalum* known to the writers is that by Nance[3] from Oklahoma in plants under the name of *Ornithogalum aureum*. The writers' material included diseased *Hyacinthus orientalis* L., variety Yellow Hammer, *Galtonia candicans* Decne., and *O. thyrsoides* from Oregon; and *Lachenalia* sp. and *O. thyrsoides* var. *aureum* Ait. from Alabama. Conspicuous mottling was noted also in leaves of red squill (*Urginea maritima* (L.) Baker) grown at the Plant Introduction Station, Glenn Dale, Maryland, from bulbs of Mediterranean origin. Mosaic is apparently widespread in a number of genera of the squill tribe (Scilleae) of the Liliaceae, but affected plants are evidently salable.

SYMPTOMS

In *Ornithogalum thyrsoides* young leaves show a fine mottling of light- and dark-green, which becomes gray or yellow and more conspicuous as the leaves mature (Fig. 1, A). Flower stalks are sometimes boldly marked with sharply contrasting light- and dark-green blotches (Fig. 1, B). In the white perianth segments, longitudinal thin streaks often appear, resembling the streaks in narcissus flowers accompanying narcissus mosaic. The mosaic patterns noted in the leaves of *Ornithogalum* and *Lachenalia* in Alabama, and in *Galtonia* and *Hyacinthus* in Oregon, were similar. In *Galtonia* grown in the greenhouse the symptoms were much milder than those noted in the open in Oregon.

TRANSMISSIBILITY

Methods

Plants for inoculation were grown from seed in a screened greenhouse kept free from sucking insects by frequent fumigations with nicotine. No natural infections in control plants were detected during the experiments of three seasons. Mechanical transfers were made by rubbing extracted sap

[1] Entomologist, Division of Truck Crop and Garden Insect Investigations, Bureau of Entomology and Plant Quarantine.

[2] Pathologist, Division of Fruit and Vegetable Crops and Diseases, Bureau of Plant Industry, Soils and Agricultural Engineering.

[3] Nance, N. W. *Ornithogalum aureum*. *In* Diseases of plants in the United States in 1939. U. S. Dept. Agr., Bur. Plant Indus., Plant Dis. Rptr. Sup. 128, p. 368. 1940. [Processed.]

on young leaves previously dusted with carborundum powder. Aphids were reared in a separate insectary greenhouse. A nonviruliferous colony of *Aphis gossypii* Glover was maintained on chrysanthemum (*Chrysanthemum hortorum* L. H. Bailey), colonies of *Myzus persicae* Sulz. and of *Macrosiphum solanifolii* Ashm. on snapdragon (*Antirrhinum majus* L.), and colonies of *Macrosiphum lilii* Monell and of *Myzus circumflexus* Buckt. on virus-free seedlings of Easter lily (*Lilium longiflorum* Thunb.).

In early experiments aphids were transferred by brush to separate infected source plants in cages, where they were left for 24 hours or, if on

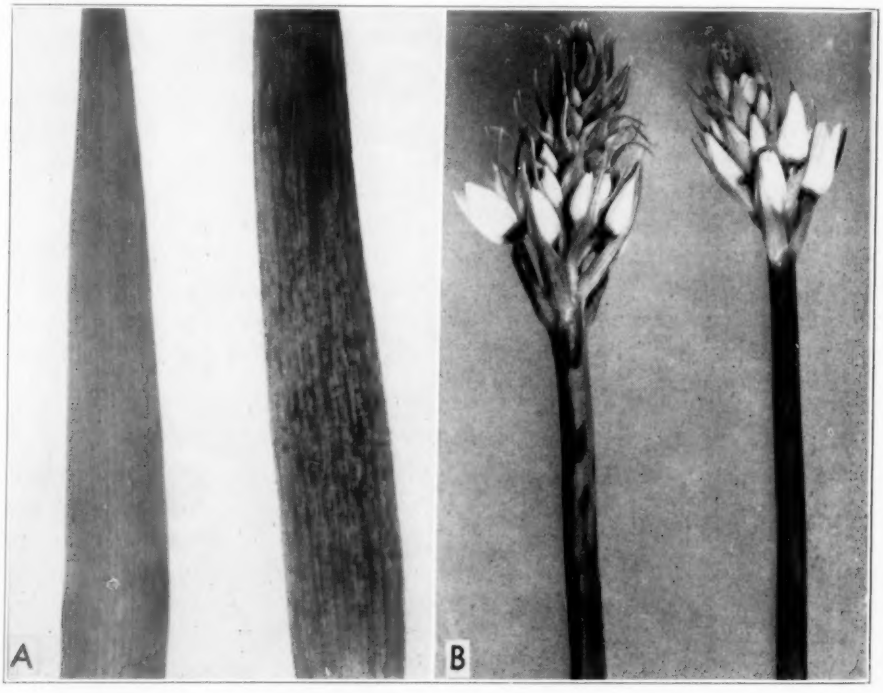

FIG. 1. Symptoms caused by Ornithogalum-mosaic virus in *Ornithogalum thyrsoides*. A. Leaf mottling, transmitted by *Myzus persicae*, in comparison with leaf of a control plant. B. Blotching of a flower stalk, transmitted by *Macrosiphum solanifolii*, in comparison with control.

favorable food plants, until needed. In later experiments the aphids were transferred to pieces of leaves taken from the source plants and placed in Petri dishes, where they fed for 1 to 3 hours. In transmission trials pieces of leaves of the source plant on which aphids were feeding were transferred to pieces of paper resting on the foliage of plants that had been brought to the insectary greenhouse and caged for this purpose. After 7 to 32 days on such test plants the aphids were destroyed by spraying or dipping in a pyrethrum preparation. The test plants were then returned to the insect-free greenhouse for observation.

Aphis gossypii fed readily and increased freely on *Ornithogalum* and

Easter lily. *Myzus persicae* from snapdragon and *M. circumflexus* from lily were readily transferred to *Ornithogalum*, where they fed on leaves and on flower parts in various stages of development. *Macrosiphum solanifolii* fed readily and established colonies on hyacinth and *Ornithogalum;* fed for several days, but failed to establish colonies on *Galtonia candicans, Agapanthus africanus* (L.) Hoffmg., and *Pancratium maritimum* L. *Macrosiphum lilii* bred continuously in large numbers on *Ornithogalum* and on Easter lily, but fed sparingly on *Zephyranthes* sp. All aphids in these tests died within 2 days on onion (*Allium cepa* L.).

Experimental Results

Experimental transfers of the virus from *Ornithogalum thyrsoides* to seedlings of this species are listed in table 1. Transfer by sap, using the carborundum-leaf-rubbing technique, proved difficult. Three of 8 trials

TABLE 1.—*Transmission of Ornithogalum mosaic virus from Ornithogalum thyrsoides to O. thyrsoides seedlings*

Method of transfer	Number of aphids per trial	Number of trials	Proportion of plants infected[a]	Minimum incubation period[b]
				Days
Mechanical	.....	8	6/75	32
Aphis gossypii	105	1	5/5	17
Macrosiphum lilii	265	1	5/5	10
Macrosiphum solanifolii	75	1	5/5	31
Myzus circumflexus	40	2	4/10	34
Myzus persicae	20–300	10	46/46	10

[a] Number of plants infected over number exposed.
[b] Interval between inoculation and appearance of symptoms.

were failures, and in the best trial only 2 of 10 plants became infected. No advantage was found in extracting the sap with phosphate buffers adjusted to pH 6, 7, or 8, or with 1 per cent dextrose or 0.85 per cent NaCl. No attempt was made to determine properties of the virus because of this low degree of success in transfer by sap.

All 5 species of aphids tested transferred the virus, and all except *Myzus circumflexus* with high efficiency. Uniform symptoms resulted from transfer by the several species of aphids and by sap, indicating that a single virus is concerned. No great importance is attached to differences in the incubation period, as daily records were not made on this point, and growth rates following inoculation varied. Under favorable conditions the symptoms may appear within 10 days following aphid transfer; much longer intervals seem to be characteristic of mechanical transfer.

The virus has been transmitted also to *Ornithogalum thyrsoides* by *Myzus persicae* from a yellow-flowered variety, *O. thyrsoides* var. *aureum*, and also from *Lachenalia*, variety Rector of Cawston, from Alabama. Symptoms typical of Ornithogalum mosaic were recognized after 18 days in each of

these 2 tests, all exposed plants becoming infected. *Myzus persicae* transferred the virus from naturally infected *Galtonia candicans* from Oregon in each of 2 trials. From mottled hyacinth variety Yellow Hammer, from Oregon, *Myzus persicae* infected 6 of 10 *Ornithogalum* plants in 1 trial. In the first subtransfer from this set to other *Ornithogalum* seedlings, 4 of 10 plants were infected; but 2 later serial transfers induced mottle in all plants exposed. This vector failed to transmit the disease from Yellow Hammer hyacinth in a second trial, and failed to transfer virus from hyacinth varieties Grand Maître and L'Innocence that showed mild mottling in leaves and flower stalks. *Myzus persicae* failed to transmit a virus from mottled red squill (*Urginea*) to *Ornithogalum* in 1 trial. *Macrosiphum solanifolii* transferred the virus from Yellow Hammer hyacinth to 1 of 5 plants of *Ornithogalum* in 1 trial. The symptoms induced in *O. thyrsoides* by the virus from *Lachenalia, Galtonia,* and *Hyacinthus* appear identical with those of the Ornithogalum mosaic virus occurring in nature.

Attempts to transfer Ornithogalum-mosaic virus to healthy seedlings of other members of the Scilleae have for the most part failed or proved inconclusive. Viruliferous *Myzus persicae* colonized on seedlings of *Eucomis undulata* Ait. produced after 6 days conspicuous angular yellow blotches in 2 of 3 plants. These supposed local lesions persisted for months, but systemic symptoms did not appear, and the virus was not returned to *Ornithogalum thyrsoides* by *M. persicae* in a trial 1 month after the original test. No symptoms whatever resulted from a second transfer to *Eucomis* under similar conditions.

No definite symptoms resulted from attempted transfers of the virus to Galtonia by *Myzus persicae* and by *Macrosiphum solanifolii,* and a subtransfer back to *O. thyrsoides* by *Myzus persicae* 2 months after exposure was also without effect. *Myzus persicae* failed to induce symptoms in seedlings of *Camassia leichtlinii* (Baker) S. Wats., *Hyacinthus azureus* (Fenzl.) Baker, *Muscari botryoides* Mill. variety Heavenly Blue, and *Scilla peruviana* L., and failed to return the virus from inoculated plants of *Camassia, Muscari,* and *Scilla.*

In other trials efficient vector species, chiefly *Myzus persicae,* failed to transmit Ornithogalum-mosaic virus to the following plants: Amaryllidaceae—*Pancratium maritimum, Zephyranthes* sp.; Iridaceae—*Tritonia crocata* (L.) Ker.; Liliaceae—*Agapanthus africanus, Allium cepa* (onion), varieties California Early Red, Utah Sweet Spanish, *A. cernuum* Roth., *A. fistulosum* L., *A. porrum* L., *Gloriosa rothschildiana* O'Brien, *Lilium formosanum* Stapf., and *L. longiflorum.* Mechanical inoculations of *L. formosanum, L. longiflorum, Tulipa gesneriana* L., variety Clara Butt, and *Nicotiana tabacum* L. variety Samsun (Turkish), resulted in no infection.

Attempts to transfer the virus from *Ornithogalum* to tulip by aphids have yielded inconsistent results. Flower breaks of types produced by tulip viruses resulted from 3 transfers of *Myzus persicae,* but not from 4 other trials. The virus was not returned from tulip to *Ornithogalum* by *M.*

persicae in either of 2 trials conducted. *Myzus circumflexus,* a proved vector of Ornithogalum mosaic virus, failed to carry this virus from *Ornithogalum* to tulip in one experiment. Inasmuch as available stocks of tulips carry some breaks, and as *M. persicae* is an efficient vector of tulip-break viruses it is possible that this species has accomplished the spread of tulip-break viruses occasionally present as contaminants, rather than the apparent transfer of Ornithogalum mosaic virus to tulip. The susceptibility of tulip to this virus must, therefore, be considered in doubt.

The following inoculations from diseased plants to *Ornithogalum thyrsoides* seedlings were without effect: Cucumber mosaic by sap (subinoculations to tobacco negative); iris mosaic by *Macrosiphum solanifolii;* mottle of Easter lily by *Myzus persicae* and also by sap (subinoculation to *Lilium formosanum* negative); Easter lily necrotic fleck complex by *Aphis gossypii* (subinoculations to tobacco and *L. formosanum* negative); narcissus white-streak[4] by *M. persicae;* and onion yellow dwarf[5] by *M. persicae.* No effect resulted from transfer of tulip virus 1, tulip virus 2, and lily latent virus from *L. formosanum* to *Ornithogalum* by *M. persicae.* The methods of transfer used have been demonstrated to be effective for the viruses concerned with the exception of that of narcissus white streak, which is not known to be carried by *M. persicae.*

Also transmissible to *Ornithogalum thyrsoides,* in addition to the virus described herein, is a recently encountered virulent form of the mottle virus of Easter lily, distinguishable from the usual mottle by the reaction of *Ornithogalum* and also by severe deforming symptoms in Easter lily. The virulent mottle virus is also transmissible by *Myzus persicae,* but induces symptoms recognizably different from those of Ornithogalum mosaic in *O. thyrsoides* as discussed in a separate publication.[6]

The virus from Yellow Hammer hyacinth, shown above to be transmissible to *Ornithogalum* by aphids, and considered identical with Ornithogalum-mosaic virus in the writers' experience, was not transmitted to Clara Butt tulips, to Easter lily seedlings, or to Croft Easter lilies by *Myzus persicae.* Mechanical inoculations from Yellow Hammer hyacinth to Clara Butt tulip, to Easter lily seedlings, to *Lilium formosanum,* and to Turkish tobacco were similarly negative. McWhorter[7] writes that the Yellow Hammer hyacinth, purchased from a commercial grower, showed characteristic mottling when first grown, and for 10 years thereafter. He was unable by mechanical methods to transfer a virus from this stock to tulip, to *Trifolium incarnatum* L., to *Vicia faba* L., or to an apparently virus-free blue-flowered variety of hyacinth. He concluded that a transitory mottling induced in some Easter lily seedlings was a shock reaction. He failed to produce any symptoms in hyacinths on inoculation with Tulip virus 1 from tulip, or with lily-mottle viruses of the tulip group from *Lilium candidum* L. and *L.*

[4] Source material from F A. Haasis.
[5] Source material from F. P. McWhorter.
[6] Brierley, Philip, and Floyd F. Smith. Studies on lily virus diseases: the mottle group. (In press.)
[7] Personal communication from F. P. McWhorter.

martagon L. He, therefore, agrees with the writers that the virus from Yellow Hammer hyacinth is not transmissible to lily or tulip, and that the usual lily and tulip viruses do not affect hyacinths.

DISCUSSION

Atanasoff[8] described mosaic symptoms in species of *Hyacinthus* and of *Muscari* that he considered due to "the mosaic disease of bulb plants," affecting various other members of the Liliaceae, Amaryllidaceae, and Iridaceae. He reported further that "the mosaic disease of tulips, hyacinths, and narcissi passes easily to narcissus and tulip." This implication that various distantly related bulbous plants are subject to a common mosaic disease has not been supported by more recent studies of Haasis[9] and of the writers.

All proved suscepts of the Ornithogalum-mosaic virus are members of the tribe Scilleae of the Liliaceae. Even within this tribe several species appear to be refractory to infection, including the Heavenly Blue variety of *Muscari botryoides*. Three species of *Muscari* were reported subject to mosaic by Atanasoff.[8] Perhaps the present failure to infect other genera of the Scilleae with this virus should be discounted in view of the difficulty of producing infection in *Galtonia candicans*, which seems subject to the same virus in nature. No explanation is evident for this difficulty of intergeneric transfer by vectors highly efficient in transfer within certain species. It was observed, however, that aphids transferred from *Ornithogalum*, a favorable host plant, to *Galtonia* failed to settle down and feed, at least for some time. It seems possible that during this interval the insects might lose the virus and fail to cause infection even though they feed later. Transfers from hyacinth Yellow Hammer to *Ornithogalum* were also erratic. A virus apparently identical with Ornithogalum-mosaic virus occurs in hyacinths, but it is uncertain whether this virus is identical with that of the hyacinth mosaic of other writers.[8]

Ornithogalum-mosaic virus shows no indication of relationship with the viruses of *Allium* or with those of the Iridaceae. Some relationship with the tulip-breaking group that causes mosaic diseases of tulip and lily is suggested. Symptoms occasionally induced in tulip on transfer from *Ornithogalum* by *Myzus persicae*, while not accepted as wholly conclusive, resemble the effects of some of the tulip-breaking group. One of the tulip-breaking group (virulent mottle of Easter lily) has been shown to affect *Ornithogalum*. On the other hand *Macrosiphum lilii* and *Myzus circumflexus*, proved vectors of Ornithogalum-mosaic virus, have not transferred viruses of the tulip-breaking group in the writers' tests. If a close relationship exists between the Ornithogalum-mosaic virus and the tulip-breaking viruses consistent transfer between tulip and Ornithogalum might be ex-

[8] Atanasoff, D. Mosaic disease of flower bulb plants. Bull. Soc. Bot. de Bulgarie 2: 51–60. 1928.

[9] Haasis, F. A. Studies on narcissus mosaic. New York (Cornell) Agr. Exp. Stat. Mem. 224. 22 pp. 1939.

pected by *M. persicae*, which is an efficient vector of both, and feeds freely on both *Ornithogalum* and tulip.

The identity of Ornithogalum-mosaic virus with the long-known but inadequately described hyacinth mosaic virus cannot be established without return inoculation to virus-free hyacinths. Virus-free hyacinths presumably can be produced from seed, but this procedure is too time-consuming to be undertaken at present. Since Ornithogalum-mosaic virus is considered sufficiently distinct from the previously known viruses to stand as a separate entity, the technical name *Marmor scillearum* is proposed, to indicate that it is known to occur only in certain members of the tribe Scilleae of the family Liliaceae.

Control of mosaic in *Galtonia, Hyacinthus, Lachenalia,* and *Ornithogalum* promises to be difficult. In view of the number of effective vector species, culture of seedlings at a distance from bulb-propagated suscepts offers most promise of success. This suggestion is supported by the fact that no evidence of seed transmission of Ornithogalum-mosaic virus was detected in 143 seedlings of *Ornithogalum thyrsoides* from mosaic-affected parents. It is of practical value to know that this common disease of minor bulbs is no menace to lilies or to onions. *Ornithogalum thyrsoides,* easily grown from seed, is of some technical interest as a test plant for viruses occurring in other monocotyledonous plants.

SUMMARY

A mosaic disease of *Ornithogalum* is described. The virus is transmissible with difficulty by sap, with a high degree of efficiency by *Aphis gossypii, Macrosiphum lilii, Macrosiphum solanifolii,* and *Myzus persicae,* and with somewhat lower efficiency by *Myzus circumflexus.* Viruses indistinguishable from this, at least in *Ornithogalum,* have been transferred from naturally infected plants of *Galtonia candicans,* hyacinth, and *Lachenalia* sp. The proved host range of the virus includes only species of these genera, and possibly of *Eucomis,* all members of the tribe Scilleae of the Liliaceae. The common name Ornithogalum-mosaic virus and the technical name **Marmor scillearum** are proposed.

DIVISION OF TRUCK CROP AND GARDEN INSECT INVESTIGATIONS,
BUREAU OF ENTOMOLOGY AND PLANT QUARANTINE,
AND
DIVISION OF FRUIT AND VEGETABLE CROPS AND DISEASES,
BUREAU OF PLANT INDUSTRY, SOILS AND AGRICULTURAL ENGINEERING,
U. S. DEPARTMENT OF AGRICULTURE.

PHYTOPATHOLOGICAL NOTES

Reaction of Lycopersicon spp. to Spotted Wilt.—Spotted wilt of tomatoes is sufficiently widespread and destructive in certain parts of the world to justify a search for resistance. Investigations by D. R. Porter (unpublished), made prior to the experiments reported herein, have established that one strain of *Lycopersicon pimpinellifolium* is resistant to spotted wilt under conditions of natural infection. Wenholz[1] has reported briefly the resistance of this species and of an unnamed Peruvian type. The following are additional observations on the resistance to spotted wilt of certain tomato species. This report is based on field infection only and plants were classified as to disease reaction solely on the basis of visible symptoms.

During the summers of 1942 and 1943, plots were located at the trial grounds of a seed company at Salinas, California, where extensive plantings of ornamentals provide an excellent source of spotted-wilt-infective thrips. In 1942 all 104 plants of various strains of *Lycopersicon esculentum* and hybrids with Porter's *L. pimpinellifolium* strain were diseased. Of 19 plants of one strain of *L. hirsutum* (P.I. 134,417), all were infected. Twenty-one plants of *L. pimpinellifolium* (Porter's strain) were tested and none showed signs of spotted wilt. Forty-eight plants of 5 strains of *L. peruvianum* (P.I. 126,930, 126,944, 126,946, 128,659 and 129,146) were planted and remained disease-free.

In 1943, 39 plants of 41 from known susceptible lots of *Lycoperiscon esculentum* and hybrids became infected. One of two selections of the German Sugar variety (*L. esculentum*), obtained from W. A. Frazier of the University of Hawaii, showed a high degree of resistance (3 out of 10 plants diseased), while the other selection was susceptible. Ten plants each of two lots of *L. pimpinellifolium*, Porter's strain, and the Fusarium-immune Accession 160 of Bohn and Tucker[2] were planted. Porter's strain remained free from spotted wilt as in previous years while 6 of the 10 plants of Accession 160 were diseased. Of 4 lots of 10 plants each of *L. peruvianum* (P.I. 126,928, 126,944, 128,657 and 128,660) one plant of P.I. 128,660 probably was diseased. The F_1 hybrid of *L. esculentum* and *L. pimpinellifolium*, Ac. 160, was susceptible, as were 5 back-cross lots of *L. esculentum* to the above hybrid. All 69 plants of these hybrids were diseased.

The 2 years' tests confirm the observations of Porter and of Wenholz regarding the resistance of *Lycopersicon pimpinellifolium*. In addition the *Peruvianum* species exhibits a high degree of resistance. The resistance of *L. peruvianum* is difficult to use because this species does not cross readily with the cultivated tomato, although this has been accomplished by an embryo culture technique (to be published elsewhere). The susceptibility

[1] Wenholz, H. Spotted wilt of tomatoes. Breeding for resistance. Hawkesbury Agr. Coll. Jour. **36**: 103. 1939. (Plant Breed. Abs. **10** (1): 65. 1940.)

[2] Bohn, G. W., and C. W. Tucker. Studies on fusarium wilt of the tomato. Immunity in *Lycopersicon pimpinellifolium* Mill. and its inheritance in hybrids. Missouri Agr. Expt. Stat. Res. Bull. 311. 1940.

of one of the two strains of *L. pimpinellifolium* shows that this species is not uniformly resistant. The resistant German Sugar variety of *L. esculentum* may have promise of hybridization. The appearance of the German Sugar suggests that this variety may have had a resistant strain of *L. pimpinellifolium* as an ancestor.—PAUL G. SMITH, Division of Truck Crops, University of California, Davis, California.

Witches' Broom of Beans.—An unreported abnormality of beans was observed at Tucson, Arizona, October 14, 1943, in a victory garden contain-

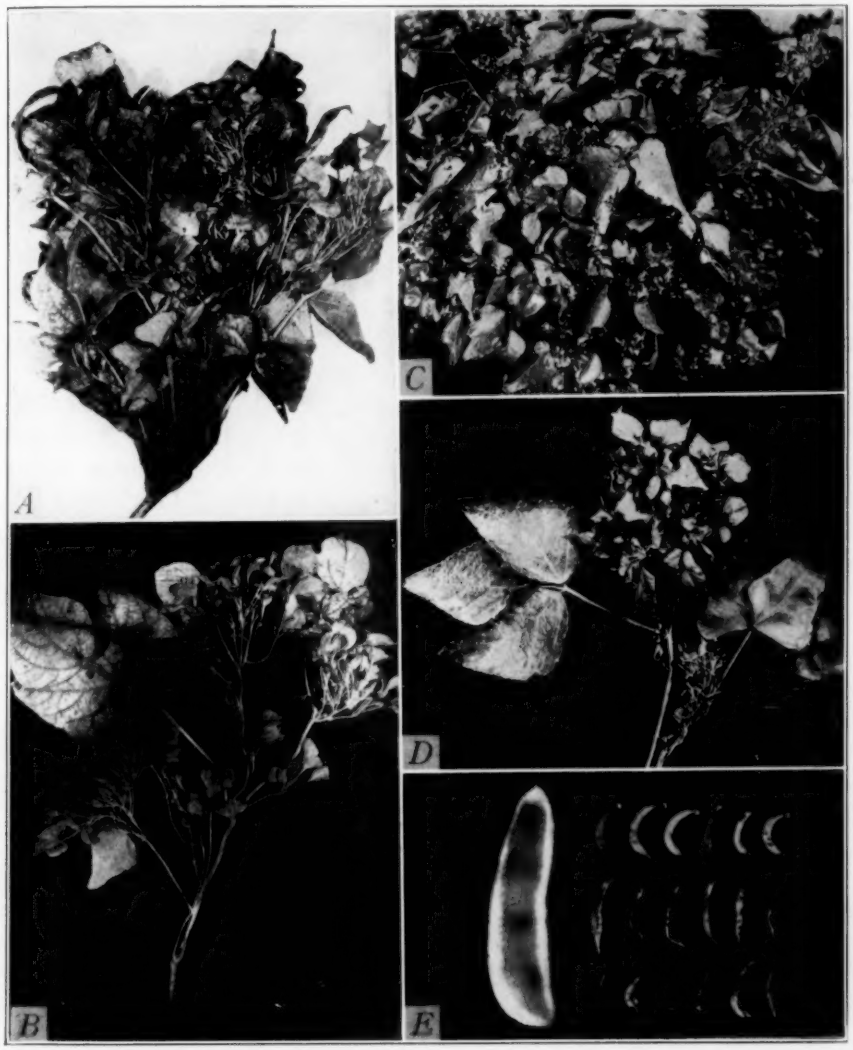

FIG. 1. A. and B. A string-bean plant and a portion thereof showing witches' broom. C and D. A Lima-bean plant and an affected portion. E. One normal and several abnormal pods. The two lower rows of affected pods show considerable wrinkling.

ing six 20-foot rows of string beans. Four plants had an excessive multiplication of branches resulting in symptoms identical to those described for witches' broom. Figure 1, A, illustrates the appearance of an affected plant, while Fig. 1, B, shows a portion of this plant. Pods on the affected parts were small and many were wrinkled. On November 18, 1943, the same abnormality was observed in another Tucson victory garden containing six 16-foot rows of Lima beans. Only one plant (Fig. 1, C) showed symptoms of witches' broom. Figure 1, D, illustrates a portion of the abnormal Lima-bean plant. The affected pods (Fig. 1, E) were $\frac{11}{16}$ to $1\frac{3}{16}$ inches long as compared to a normal pod (Fig. 1, E) measuring $3\frac{11}{16}$ inches. Approximately two-thirds of the affected pods were wrinkled.

Climatic conditions at certain locations in southern Arizona are favorable for spring and fall crops of beans. According to the owners of the victory gardens, their spring beans did not show symptoms of witches' broom. In case of the string beans, the seed for the spring and fall plantings were taken from the same package.—WILLIAM G. HOYMAN, Emergency Plant Disease Prevention Project, Department of Plant Pathology, University of Arizona, Tucson, Arizona.

The Perennial Tree Onion a Carrier of Onion-Yellow-Dwarf Virus.—The perennial tree, top, or topset onion (*Allium cepa* L. var. *viviparum* Metz.) is grown rather widely through the northern tier of States, especially in home gardens, as an early green onion. It is found frequently in gardens in the area of commercial-onion culture in New York State, and is occasionally grown as a market-garden crop in the North, in fields of an acre or more. Newhall[1] has shown that this variety is a factor in overwintering of onion mildew (*Peronospora destructor* (Berk.) Caspary). Circumstantial evidence suggested to H. A. Jones that the tree onion was concerned also in the overwintering of onion-yellow-dwarf virus (*Allium virus 1* (Melhus) Smith, *Marmor cepae* Holmes). Accordingly, 13 plants from New York were supplied to the writers in December, 1942, for the purpose of determining the presence of virus and the symptom expression of onion-yellow-dwarf virus in this variety.

Planted in pots in a greenhouse on December 7, 1942, 4 of 13 plants showed mild yellow streaking at the bases of young leaves on January 1, 1943, but no further confirming symptoms developed on continued growth through November 17, 1943. *Myzus persicae* (Sulz.), fed on leaves of 5 tree onions and transferred to 5 California Early Red seedlings on December 24, 1942, induced strong typical yellow-dwarf symptoms in 1 of 5 test seedlings, recognizable after 14 days and persisting thereafter. On the same date, yellow-dwarf virus from Oregon was inoculated into 5 tree onions by *M. persicae*. No symptoms sufficient for diagnosis appeared up to March 1, 1943, when the same 5 plants were reexposed by *M. persicae* to yellow-dwarf virus out of multiplier onion (*Allium cepa.* var. *solaninum* Alef.), from

[1] Newhall, A. G. The spread of onion mildew by wind-borne conidia of *Peronospora destructor*. Phytopath. 28: 257–269. 1938.

West Virginia. No reliable symptom expression was detected on continued growth to October 20, 1943, when the 5 plants twice exposed to yellow-dwarf by aphids and the 8 plants not experimentally exposed were individually indexed by the carborundum leaf-rubbing method, 5 California Early Red sets serving as test plants for each tree onion inoculation.

These tests showed 6 of 8 noninoculated tree onions infected with yellow-dwarf virus, as well as 5 of 5 that had been inoculated by aphids. In the tests of tree onions that proved positive, from 1 to 5 of the inoculated Early Red sets were infected, while a parallel transfer of yellow-dwarf virus from West Virginia multiplier onions to sets of the same lot by the same method produced 148 infections in 150 sets inoculated. No yellow-dwarf has appeared in control seedlings or sets during 2 seasons' tests. The lower proportion of transfer from the tree onion may indicate that the virus concentration in this tolerant variety is lower than in less tolerant onions, but no evidence was found that the New York virus from tree onion was less virulent than the West Virginia virus from multiplier onion. Two tree onions yielded no virus in these index trials. If repeated indexing reveals no virus in these 2 plants they will serve to produce a virus-free stock of this variety.

The tree onion is essentially a symptomless carrier of yellow-dwarf virus, for no symptoms sufficient for accurate diagnosis were detected during 11 months' growth in the greenhouse in plants thus shown to be infected. The multiplier onion, also grown as a winter onion in gardens, expressed typical yellow-dwarf symptoms in 7 of 12 plants received from West Virginia in 1942. Both varieties of winter onions, the tree and the multiplier, are capable of overwintering the yellow-dwarf virus. In the multiplier variety diagnosis can be made as readily as in commercial varieties, but this is not the case with the tree onion.—PHILIP BRIERLEY and FLOYD F. SMITH, Plant Industry Station, Beltsville, Md.

A Simplified Method of Growing Plants with Roots in Nutrient Vapors.—To facilitate studies[1, 2] of Phytophthora infections and of the toxicity of various concentrations of nitrite and other ions on citrus and avocado roots, the apparatus shown in figure 1 was assembled. This consists of a 12-liter cylindrical glass jar containing 3 liters or less of nutrient, which is recirculated in vapor form over the suspended roots by means of a DeVilbiss atomizer operated with air pressure. To the intake of the atomizer is attached a glass tube, which extends to the bottom of the jar. Clogging of the atomizer by sloughed root material is prevented by filtering the nutrient entering the intake through Pyrex glass wool.

Several obvious advantages of the method are that it permits easy inoculation and examination of the roots, supplies abundant aeration, and readily allows the use of any number of cultures in separate containers of any suit-

[1] Klotz, L. J., and V. P. Sokoloff. The possible relation of injury and death of small roots to decline and collapse of citrus and avocado. Citrus Leaves **23**: 1, 2, 3, and 22. 1943.

[2] Sokoloff, V. P., and L. J. Klotz. Distress of citrus trees in association with the temporary presence of nitrite in soil. Citrus Leaves **23**: 8–10. 1943.

able size. It also obviates the disadvantage of dilution[3] because the nutrient is recirculated. One needs only to replace occasionally the small amount of solution that escapes as vapor through the holes of the plant support. If desired, the solution level can be maintained automatically with a fount-type of supply vessel. The set-up also lends itself readily to use with the con-

FIG. 1. Apparatus for growing plants with roots in vaporized nutrient.

tinuous flow method[4, 5] of supplying nutrient.—L. J. KLOTZ, Division of Plant Pathology, University of California Citrus Experiment Station, Riverside, California.

A Note on the Effects of Splash Injury in Guayule Seedlings.—Guayule seedlings have been grown customarily in nurseries employing overhead

[3] Carter, Walter. A method of growing plants in water vapor to facilitate examination of roots. Phytopath. 32: 623–625. 1942.

[4] Chapman, H. D., and George F. Liebig, Jr. Adaptation and use of automatically operated sand-culture equipment. Jour. Agr. Res. **56**: 73–80. 1938.

[5] Eaton, Frank M. Automatically operated sand-culture equipment. Jour. Agr. Res. 53: 433–444. 1936.

irrigation systems. This method of watering sometimes produced an injury resulting in high loss of young seedlings in the cotyledon stage. For want of better name this has been called "splash injury." A brief investigation

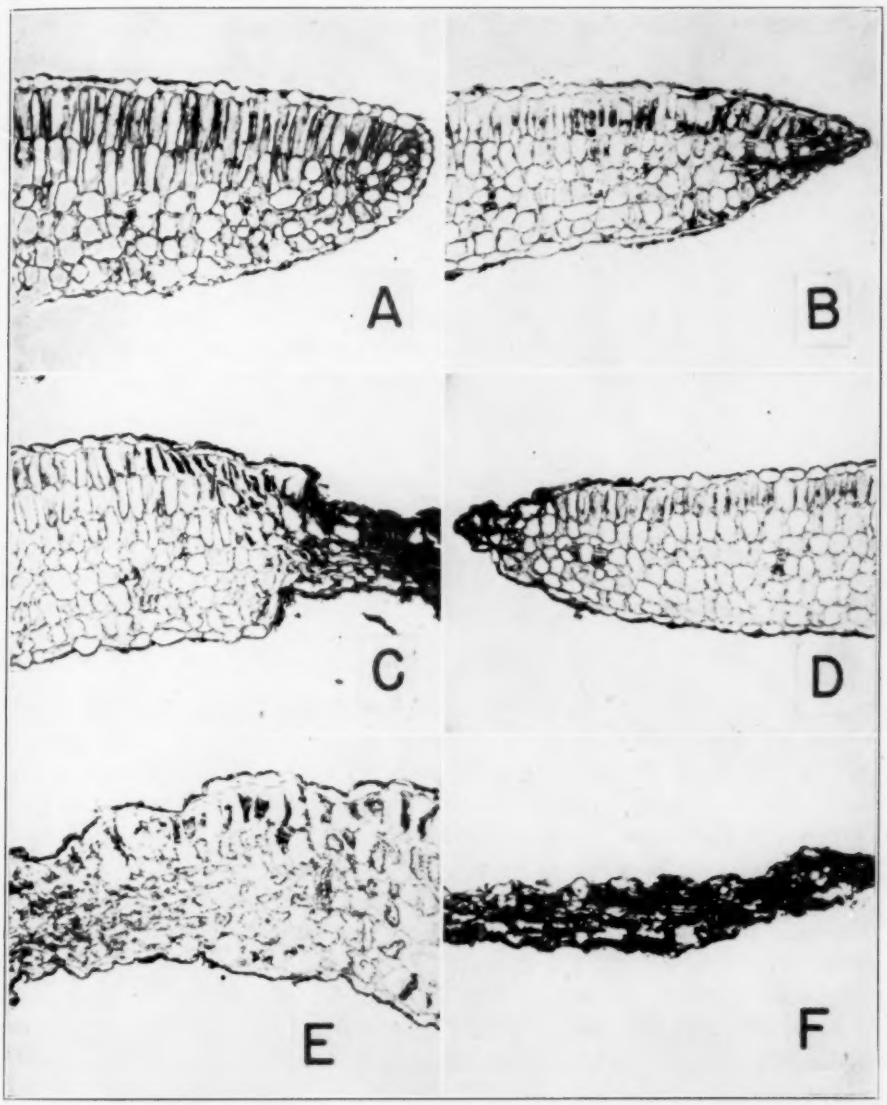

FIG. 1. Sections of splash-injured guayule cotyledons. 1, A. Uninjured cotyledon. 1, B–F. Various degrees of injury. Magnification about 75 ×.

was undertaken to determine (a) the anatomical nature of the injury, and (b) whether microorganisms were present in the injured tissues.

Seedlings showing various degrees of injury were collected from the Alisal Nursery at Salinas, California. They were fixed in form-propriono-

ethanol, dehydrated by means of tertiary butyl alcohol, and stained by the safranine-fast green method of Johansen and the differential fungus-host stains of Ikata and Margolena. The above methods were carried out according to the directions of Johansen's *Plant Microtechnique*. Observations were restricted to sections of cotyledons, as little of the material collected showed injury below the cotyledonary node.

External symptoms of irrigation-splash injury are principally a brown discoloration and withering at the tips and edges of the cotyledons. In severe cases the entire cotyledon, as well as the hypocotyl, are affected.

Various degrees of injury can be seen in the photomicrographs of sections of cotyledons. A portion of an uninjured cotyledon is shown in figure 1, A. Injury usually appeared around the edges of the cotyledon first (Fig. 1, B, D) and then progressed inwards. It sometimes involved the entire cotyledon more or less simultaneously (Fig. 1, E). In either case the final result was the complete collapse of practically all of the parenchyma cells in the cotyledon (Fig. 1, F).

The collapse of cells is preceded by plasmolysis (Fig. 1, E, F) and cytolysis (Fig. 1, B, C, E).

No evidence of microorganisms was observed in the cotyledon sections or in the cotyledonary node and hypocotyl sections. The immediate cause of the injury is, therefore, presumed to be nonpathogenic, brought about in some manner, either by the soil or by water striking the plant or accumulating around it.—FREDRICK T. ADDICOTT, Guayule Research Project, Salinas, California.

A Severe Necrosis Caused by Bean-Mosaic Virus 4 on Beans.—It was shown that bean mosaic virus 4 (*Marmor laesiofaciens* Zaum. and Harter) causes only local infection on some varieties and only systemic infection on others, manifested by a leaf mottling.[1] The varieties susceptible to local infection are immune from systemic infection, and the systemic infected varieties show no local lesions.

A third type of response was recently noted in a strain of the Blue Lake variety. Instead of the local lesions that are produced by the virus on most strains of the variety (Fig. 1, A), small, sometimes indistinct necrotic lesions were observed on the inoculated leaves, mostly in the region of the veins and veinlets (Fig. 1, B). These varied from small, pin-point, dark spots to slightly larger necrotic areas. In some cases only a few lesions were visible, whereas in other cases they were fairly numerous and well-distributed over the entire inoculated leaf. This effect was followed in some instances by a veinal necrosis of varying degrees, depending upon the extent of the infection. Such leaves frequently were killed more rapidly than similar leaves on which only local lesions were produced. A similar type of response was

[1] Zaumeyer, W. J., and L. L. Harter. Two new virus diseases of beans. Jour. Agr. Res. [U. S.] 67: 305–328. 1943.

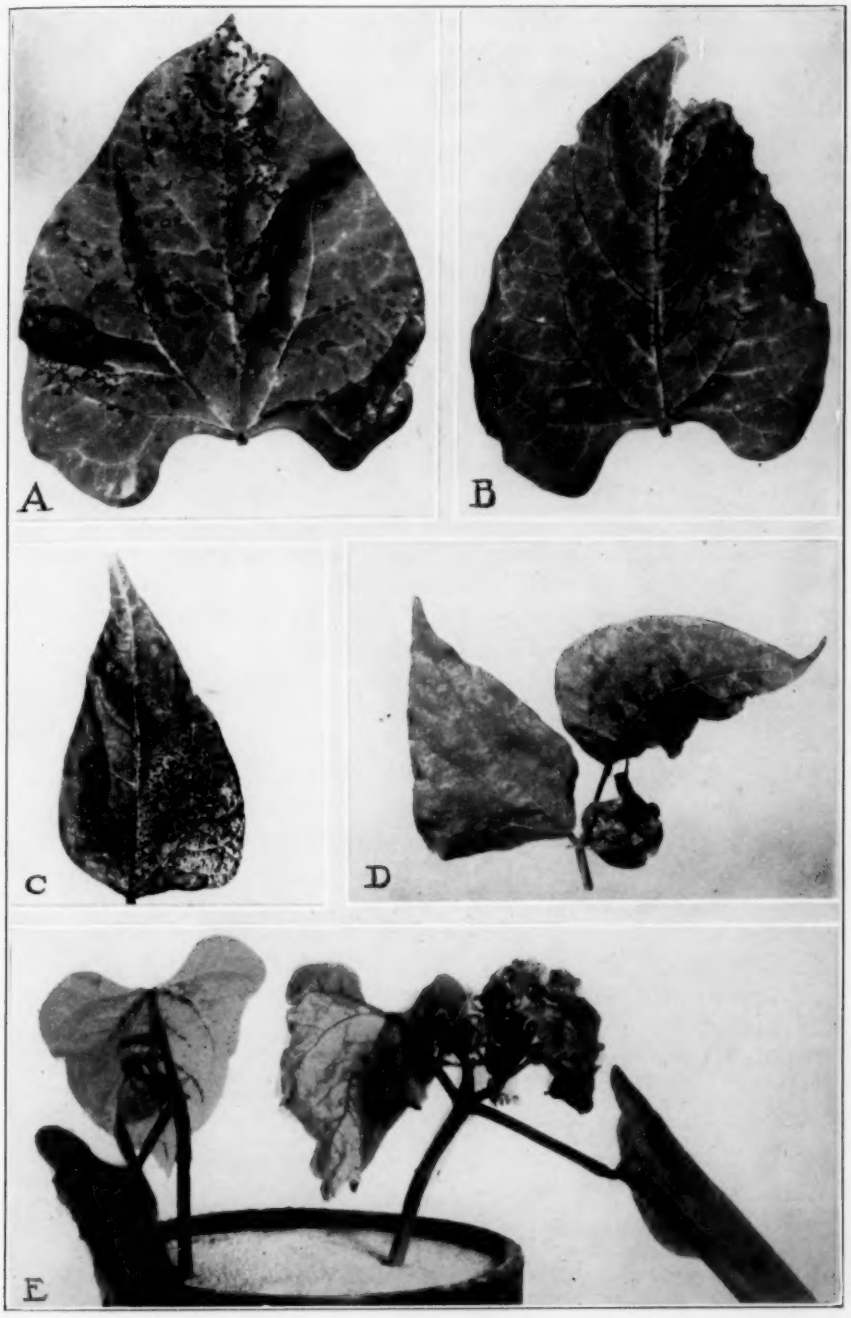

FIG. 1. Variations in symptoms produced by bean mosaic virus 4 on several varieties. A. Local lesions on Pinto. B–E. Effects on a strain of Blue Lake. B, C, necrosis of a primary and a trifoliate leaflet, respectively; D, systemic infection of trifoliate leaflets; E, rosetting of trifoliate leaflets and discoloration of stems and petioles.

reported by Holmes[2] in a few varieties of pepper infected with the ordinary tobacco mosaic virus (*Marmor tabaci* Holmes).

About 10–12 days following these symptoms, irregularly shaped, reddish, necrotic spots appear, most frequently on the upper side of one or more of the trifoliate leaflets (Fig. 1, C). On the underside, the veinlets become reddened, although this effect is frequently noticeable on the upperside as well. Another characteristic symptom is a drooping of the infected leaflets at the pulvini. Although an entire leaflet may show this necrosis, it is common to note the infection on only one side of the midrib (Fig. 1, D). Growth may be almost completely stopped on the necrotic side and thus the leaflet becomes distorted and curls toward the affected portion (Fig. 1, D). In some instances only one leaflet is so affected, whereas in other cases similar symptoms develop on all of the leaflets. Infected leaflets may die, while others may be severely malformed, puckered, and much smaller in size than normal ones (Fig. 1, D).

The stems and petioles of infected plants always show a darkening or a dark, reddish streaking, followed by a slight shrinking.

Another, although less common, type of symptom is characterized by the failure of the young trifoliate leaves to develop, or else when they do develop they become extremely rosetted (Fig. 1, E). They are chlorotic, thickened, and brittle. The internodes are very short, somewhat shrunken, darkened, and extremely brittle. The internal tissues also are necrotic. Such plants frequently remain in this condition for several weeks before dying.

Usually, those plants that showed the greatest amount of necrosis on the inoculated primary leaves were the first to show the systemic necrosis on the trifoliate leaves. Certain inoculated plants that did not show necrotic spots on the primary leaves, manifested a less severe type of necrosis on the trifoliate leaves, which appeared later than on plants developing the necrosis on the primary leaves.—W. J. ZAUMEYER and L. L. HARTER, Division of Fruit and Vegetable Crops and Diseases, Bureau of Plant Industry, Soils, and Agricultural Engineering, Agricultural Research Administration, Plant Industry Station, Beltsville, Md.

[2] Holmes, F. O. Inheritance of resistance to tobacco mosaic disease in the pepper. Phytopath. 27: 637–642. 1937.

BOOK REVIEW

NAUMOV, N. A. *Rzhavchina Khlebnykh Zlakov v SSSR.* [*Rusts of Cereals in USSR*]. Gosud. Izdat. Kolkh. i Sovkh. Liter., Moscow and Leningrad. 401 pp. 64 figs., 1939. 18r. 50 k. (With chapters on varietal resistance to rusts, selection for rust resistance, and control by agrotechnical methods by E. E. Geshele, and one on damage from rusts by A. A. Shitikova-Rusakova.)

That cereal rusts are no less a problem in the Soviet Union than "in capitalistic countries" is evidenced by such rust losses as 24 per cent (111,000,000 bushels) of the wheat crop and 26 per cent (140,000,000 bushels) of the oats crop in 1933 in but two of the many cereal-producing provinces in Russia. Recognition of the importance of this problem led to the regimentation of Russian phytopathologists and agronomists, beginning some 15 years ago, in an integrated program of developing means of rust prevention. Throughout the past decade the cereal rust literature has been enriched by an increasing influx of practical and theoretical contributions from the Russian workers, many of them original, stimulating, and valuable.

Naumov's "Rusts of Cereals in USSR," the first comprehensive work of this character since that of Yachevski, 35 years ago, comes as a welcome introduction and guide to this new pathological activity. With its background, it is to be expected that the prevailing emphasis of the book is on the practical problem of rust control, yet the theoretical bases of rust etiology and control are not neglected.

Following three chapters dealing with the practical significance of rusts, their developmental cycles, and the significance of heterothallism, there is an extended treatment of the nature and methods of estimation of rust damage, a field in which Naumov, Shevchenko, Rusakov and his coworkers, Ruzinov, and other Russians have made notable, though not well-known contributions.

The following six chapters deal with the nature of rust resistance, varietal susceptibility of cereals to rust, and relation to environment, specialization, and mycology of the cereal rust fungi. *Puccinia graminis, P. triticina, P. glumarum,* and *P. coronifera* are individually treated in detail, while *P. dispersa* and *P. anomala* are touched on briefly. In Russia the most destructive of these is considered to be *Puccinia triticina,* the leaf rust of wheat. On page 33, the author states: "Brown rust (in comparison with stem and stripe rusts) occupies a much wider range in Russia, is better adapted to the climate, appears earlier, develops more quickly, and isn't afraid of the weather," while (p. 204) "as for its economic significance, this species of rust is considered the most important and brings about the most substantial damage to wheat in all the regions where it is met with. In this regard the importance of *P. triticina* is dominant, even over that of *P. graminis,* if we do not count relatively few regions where the economic significance of the two species is locally reversed."

Thirty pages are devoted to epiphytology, another phase in which the Russians, in particular Stepanov, have made noteworthy contributions. Features of cereal rust epiphytology in Russia include: relative unimportance of alternate hosts except locally; long-distance air transport of urediospores, as from Manchuria to Amur province in Siberia; survival of rusts in winter grain fields where the latter are protected by snow cover, forest strips, or features of the microrelief; and local dissemination from winter to spring cereals.

Species of *Thalictrum* play no part in the epiphytology of wheat leaf rust in Russia. The author recounts the work of Brizgalova, who has shown that in Eastern Siberia *Isopyrum fumarioides* is a functioning intermediate host for this rust. On the basis of this work, *Puccinia triticina* is divided into two varieties, var. *isopyri* for the East Siberian form and var. *tritici* for the fungus as known elsewhere in the world. The variety *isopyri* is based on its specialization on *Isopyrum,* its limited geographic range, and the early production and extended period of maturity of its teleutospores. Since the reaction of "*P. triticina* var. *tritici*" on *Isopyrum* has not been tested, the behavior of the teleutospores may be merely a reaction of the typical fungus to Eastern Siberia's rigorous climate, and geographic range alone is not a valid basis for varietal distinctions, it appears to the reviewer that this varietal separation lacks experimental verification.

In control of cereal rusts in Russia, the greatest emphasis is laid on breeding for rust resistance and agrotechnical (cultural) methods. Eradication of alternate hosts is considered to be of local importance only, and sulphur dusting, which has been the subject of study by a number of workers (Shitikova-Rusakova, Rusakov, Zaitseva and Popova, Brizgalova) has not developed to a point of economic significance. Considerable success has been achieved in breeding and selection. The work is carried out under specific instructions of the All-Russian Institute of Plant Husbandry, which includes a formal progression of new strains from breeding nursery to selection nursery, control experiments, preliminary (yield and quality) tests, and, finally, competitive tests at various stations. In the first two nurseries, selection for rust resistance is by observation only, in the last

three, accurate measurements of rust are employed. Breeding and selection have resulted in the production of a number of rust-resistant high-yielding, and adapted strains, including Zarya, and selections from *Triticum erythrospermum, ferrugineum, caesium,* and *lutescens* and from the American wheats Marquis, Kanred, Kanred × Fulcaster, and Illini Chief.

Rust control by cultural methods, in addition to practices common in the United States, includes isolation of fields of winter cereals, early spring harrowing of winter cereals, and vernalization of spring-cereal seed (which favors rust-escape by advancing the date of maturity).

Finally, two chapters are devoted to techniques of working with rusts in the laboratory, field, and nursery.

The extensive bibliography includes approximately 600 Russian and 1250 non-Russian titles, and is useful as a check list; however, it is poorly correlated with the text. A high percentage of the works referred to by author and year in the text cannot be found in the bibliography, and many of these represent important bibliographic omissions. An additional difficulty is illustrated by the reference on page 232 to "Gorlenko, 1934." There are 5 papers by Gorlenko in 1934 listed in the bibliography; this problem recurs frequently. There is a 4-page index.

The book is printed on a grade of paper resembling news print, and in a few cases the type is poorly legible; however, the illustrations, a majority of which are from American works, are sufficiently clear. It does not appear possible to purchase the book in the United States, but it may be ordered from Russia through the Four Continent Book Corp., 253 Fifth Ave., New York, N. Y. A translation of sections on epiphytology is available in the libraries of the U. S. Dept. of Agriculture, the University of Minnesota, and Oklahoma Agricultural and Mechanical College.—K. STARR CHESTER, Oklahoma Agricultural and Mechanical College, Stillwater, Okla.

REPORT OF THE 35TH ANNUAL MEETING AND WAR CONFERENCE OF THE AMERICAN PHYTOPATHOLOGICAL SOCIETY

The 35th annual meeting and war conference of The American Phytopathological Society was held at The Neil House, Columbus, Ohio, from December 4 to 6, 1943. Fifty-eight papers were accepted by the editorial committee and presented at the meeting. Outstanding was the general interest in all of the sessions and the brisk discussions following presentation of the papers and during special conferences. Approximately 200 members attended the meeting.

The Phytopathologists' dinner was held Sunday evening, December 5, at The Neil House. A delightful and informal program was enjoyed.

The special conferences and discussions included a "fungicide discussion session," the object of which was to discuss the problems of copper and organic fungicides. A round-table conference on the "emergency plant disease program" included progress reports and discussions of needed research and extension based on results of the surveys and more accurate measurements of plant disease prevalence and losses. A report of "insect pest surveys" of the Bureau of Entomology and Plant Quarantine was also given. A symposium of "vegetable seed-borne diseases" was held with discussion centered around the changing scene of vegetable seed production as influenced by the unprecedented demands brought about by lend-lease requirements and Victory gardens. A special session was devoted to the activities of the "war committee." This discussion included the history of the war committee and the present major objectives. This was followed by reports of regional committees and sub-committees of the war committee with considerable discussion from the floor. A conference sponsored by the "extension work and relations committee" included an open-informal discussion on the "possibilities for the plant pathologists to increasingly aid in the war effort," and "how the extension work and relations committee could help the pathologists in 1944." A conference sponsored by the Special Committee "Coordination in cereal and vegetable seed treatment research" included summaries by the various sub-committees plus discussions and suggestions on seed treatments of vegetables, corn, small grains, forage and pasture crops.

The 1943 annual meeting and war conference was considered a very successful meeting and the Society recommended that the "program committee" schedule its 1944 annual meeting at approximately the same time of year.

OFFICERS, REPRESENTATIVES, AND COMMITTEES FOR 1944

Officers:

J. J. CHRISTENSEN, President (1 yr.), University Farm, St. Paul, Minnesota.
J. B. KENDRICK, Vice-President (1 yr.), University Farm, Davis, California.
C. C. ALLISON, Secretary (3 yr. term expires 1944), The Ohio State University, Columbus, Ohio.
R. M. CALDWELL, Treasurer (3 yr. term expires 1946), Purdue University, West Lafayette, Indiana.

Councilors:

R. S. KIRBY (term expires 1945), Pennsylvania State College, State College, Pa.
J. G. LEACH (term expires 1944), University of West Virginia, Morgantown, West Virginia.
J. C. WALKER (term expires 1944), University of Wisconsin, Madison, Wisconsin.
H. P. BARSS (Potomac Division), Office of Experiment Stations, U.S.D.A., Washington, D. C.
O. C. BOYD (New England Division), Massachusetts State College, Amherst, Massachusetts.
A. G. PLAKIDAS (Southern Division), Louisiana State University, Baton Rouge, Louisiana.
E. E. WILSON (Pacific Division), University Farm, Davis, California.

Representatives:

A.A.A.S. Council. L. M. HUTCHINS, J. G. LEACH.
Division of Biology and Agriculture, National Research Council. J. C. WALKER (3 yr. term expires June 30, 1946).
Board of Editors, American Journal of Botany. A. A. DUNLAP (3 yr. term expires 1946).

Standing Committees:

Donations and Legacies. J. G. BROWN, N. J. GIDDINGS, B. L. RICHARDS, N. E. STEVENS, R. P. WHITE, Chm.

Extension Work and Relations. R. J. HASKELL, G. W. KEITT, J. O. MILLER, R. H. PORTER, OTTO REINKING, R. C. ROSE, D. R. SANDS, W. B. TISDALE, O. D. BURKE, Chm.

Investments. MARVIN E. FOWLER, L. M. HUTCHINS, J. W. ROBERTS, R. M. CALDWELL, Chm.

Necrology. M. B. WAITE, A. G. JOHNSON, Chm.

Phytopathological Classics. H. H. WHETZEL, Mgr., H. B. HUMPHREY, Editor.

Public Relations. L. M. BLANK, O. C. BOYD, K. STARR CHESTER, C. T. GREGORY, J. H. JENSEN, FRANK MCWHORTER, A. G. NEWHALL, J. A. PINCKARD, C. S. REDDY, LUTHER SHAW, G. H. STARR, C. E. YARWOOD, G. F. WEBER, Chm.

Recognition of Merit. CHARLES CHUPP, J. G. LEACH, L. M. HUTCHINS, J. C. WALKER, C. R. ORTON, Chm.

Regulatory Work and Foreign Plant Diseases. C. R. ORTON, R. P. WHITE, E. C. STAKMAN, Chm.

Union of American Biological Societies (and Biological Abstracts)—5 yr. term. C. W. BENNETT, DONALD FOLSOM, L. M. MASSEY, W. G. STOVER; C. C. ALLISON and HELEN HART (ex officio), F. V. RAND, Chm.

Special Committees:

Coordination in Cereal and Vegetable Seed Treatment Research. C. H. ARNDT, F. J. GREANEY, C. M. HAENSELER, K. W. KREITLOW, L. D. LEACH, R. W. LEUKEL, J. H. MCLAUGHLIN, GEORGE SEMENIUK, M. B. MOORE, Chm.

Fungus Nomenclature. C. M. TUCKER, D. S. WELCH, ERDMAN WEST, G. L. ZUNDEL, J. A. STEVENSON, Chm.

Nomenclature and Classification of Plant Viruses. C. W. BENNETT, E. W. BODINE, EUBANKS CARSNER, JAMES JOHNSON, H. H. MCKINNEY, FRANK MCWHORTER, FREEMAN WEISS, Chm.

Publication Problems. MAX GARDNER, FRANCIS O. HOLMES, A. J. RIKER, Chm.; R. M. CALDWELL and HELEN HART (ex officio).

Reorganization of International Cooperation. H. P. BARSS, G. H. COONS, J. G. HARRAR, OTTO REINKING, J. A. STEVENSON, E. C. STAKMAN, Chm.

Standardization of Fungicidal Tests. M. C. GOLDSWORTHY, C. S. HOLTON, J. G. HORSFALL, R. W. LEUKEL, C. F. TAYLOR, H. W. THURSTON, J. D. WILSON, S. E. A. MCCALLAN, Chm.

Terminology (Nomenclature) of Immunology and Use of Technical Words. JESSIE I. WOOD, N. E. STEVENS, Chm.

War Committee. I. E. MELHUS, E. C. STAKMAN, J. G. LEACH, Chm. (Executive Committee).

Temporary Committees for 1943:

Auditing. H. A. RODENHISER, S. P. DOOLITTLE, Chm.

Resolutions. CHARLES CHUPP, I. E. MELHUS, C. M. TUCKER, Chm.

Society Organization. GEORGE ARMSTRONG, R. S. KIRBY, L. D. LEACH, R. M. CALDWELL, Chm.

REPORTS OF OFFICERS, REPRESENTATIVES AND COMMITTEES FOR 1943

Report of the Secretary. At the time of our Council Meeting, February 12, 1943, the membership was 1086. It now totals (December 6, 1943) 1060. This makes a net loss of 26 members. During the period from February 12 to December 6, 1943, 30 individuals applied for membership. By action of the Society all applicants were elected on December 6.

Eight former members have been reinstated, and the Society lost a total of 64 members: 11 by resignation, 3 by death, and 50 suspended for nonpayment of dues. Of the full membership, 155 are paid-up life members, and one was paying $10.00 a year toward life membership until his payments were interrupted by conditions abroad.

The Secretary calls attention to the fact that suspended members may be reinstated by notifying the Secretary and paying dues for the current year. Former members should be advised of this and urged to seek reinstatement, if their professional work is directly related to plant diseases and their control.

The Clearing Agency of the Society was continued in 1943. There are 71 applications in the active file—27 of these new during the year. One hundred three applications of 51 pathologists were sent to 14 employers, who made request for applications. Several applicants were hired by contacts made through our Clearing Agency.

Report of the Treasurer. Statement of accounts for the year ending November 30, 1943.

Receipts:

Balance from 1942			$1964.28
Annual dues:			
1939–1940	$ 10.00		
1941	10.00		
1942	46.42		
1943	2766.41		
1944	1926.90		
1945	1.00	$4760.73	
Voluntary dues		5.00	
Donation for foreign subscription		5.00	
30-Year Index		6.25	
Sales		1.00	
Reimbursement for checks returned by bank		9.50	
Total receipts			4787.48
			$6751.76

Expenditures:

Member subscriptions transferred to PHYTOPATHOLOGY:			
1939–1940	$ 8.00		
1941	8.00		
1942	36.67		
1943	3554.16	$3606.83	
Transferred to PHYTOPATHOLOGY for:			
Voluntary dues	5.00		
Foreign subscription donation	5.00		
Sales	1.00		
Index	6.25	17.25	
Transferred to Sinking Fund		15.00	
Secretarial work and expenses of office of Secretary		215.15	
Secretarial work for Treasurer		288.00	
Printing		208.73	
Stamps and envelopes		63.39	
Express		0.63	
Demonstration program		6.96	
Bank service charge		13.54	
Checks returned by bank		20.00	
Total expenditures			4455.48
Balance on hand			2296.28
			$6751.76

Sinking Fund. The Sinking Fund, the income from which is used for the support of PHYTOPATHOLOGY, was obtained by deposit of payments from patron memberships and deductions from life-sustaining membership instalments. All life-sustaining members, with the exception of one in Italy, have paid in full. There has been no change in the amount of the fund during the past year, the total remaining at $9676.00. It is invested as follows:

First mortgage note deposited with McLachlen Banking Corporation for collection ($500.00 at 4½%)	$ 500.00
United States Savings Bond, Series G, 2½%	1000.00
Invested with the following building and loan associations:	
Arlington and Fairfax Bldg. and Loan Association, 4%	1000.00
Columbia Permanent Building Association, 4%	520.20
District Building and Loan Association, 3½%	1556.78
National Permanent Building Association, 4½%	2091.00
Northwestern Federal Savings and Loan Association, 3½%	2000.00
Perpetual Building Association, 4%	1040.40
Prudential Building Association, 3½% (accrued interest, $14.71)	190.71
	$9899.09
Less interest due PHYTOPATHOLOGY	223.09
	$9676.00

The Lyman Memorial Fund, obtained from voluntary contributions, now totals $3175.32. The whole amount is invested with the Brookland Building Association, at 3½%. The account for 1943 follows:

Balance on hand, December 1, 1942	$3132.57
Dividends, December 31, 1942, and June 30, 1943	110.51
Contributions from members	37.00
Sale of Erwin F. Smith Memoir	5.75
	$3285.83
Less interest due PHYTOPATHOLOGY	110.51
	$3175.32

Additional Endowment. During the past year the Committee on Legacies and Donations has secured as of this date (Dec. 1, 1943) and turned over to the Treasurer, United States Savings Bonds, Series F, having maturity value of $725, and stamps and cash totaling $6.50.

Report of the Business Manager. At the close of 1942 there were 416 nonmember subscribers, including 6 complimentary. In 1943 there were 7 cancellations and 48 suspensions for nonpayment, a loss of 55. As there were 70 new or restored subscriptions, a slight net gain of 15 resulted, bringing the total subscription list to 431. The gain from 200 to 240 in the domestic list was due in large part to the fact that 24 subscriptions for eventual shipment to U. S. S. R. were ordered sent to their agent in New York. There was also an order for 7 subscriptions to be sent to New York, for storage until the numbers can be sent to The Netherlands. There were a few other orders for subscriptions for future shipment to foreign countries. Again the American Library Association has ordered and paid for 45 sets of Vol. 33, to be stored with our printers for eventual shipment to foreign countries as they may designate. These 45 are not included in the total subscriptions above, but are listed as sales.

Statement of accounts for the year ending November 30, 1943.

Receipts:		
Balance from 1942		$ 7010.09
Subscriptions:		
1937	$ 6.50	
1939–1941 incl.	19.50	
1942	192.85	
1943	2214.50	
1944	482.35	
1945	17.00	$2932.70
Member subscriptions:		
1939–1940	8.00	
1941	8.00	
1942	36.67	
1943	3554.16	3606.83
Sales of back numbers (including orders for current numbers reserved for American Library Association)		999.70
Advertising:		
1942	289.41	
1943	834.33	1123.74
30-Year Index		227.60
Interest on Sinking Fund:		
First mortgages	88.40	
Building and Loan*	110.10	198.50
Interest on current funds		158.88
Grant from Rockefeller Institute		600.00
Allowance on reprints		466.93
From authors for excess illustrations		221.54
Part payment for article in November, 1942, issue		50.00
First mortgage note paid in full		1000.00
Total receipts		11586.42
		$18596.51

* $223.09 accrued interest left on deposit with Building and Loan Associations, $110.51 with Lyman Fund.

Expenditures:

Printing, distributing, and storing PHYTOPATHOLOGY:			
Vol. XXXII, No. 11	$900.78		
No. 12 and Index	754.54		
Vol. XXXIII, No. 1	734.07		
No. 2	726.77		
No. 3	704.75		
No. 4	673.58		
No. 5	721.47		
No. 6	836.45		
No. 7	857.57		
No. 8	904.29		
No. 9	719.24		
No. 10	996.70	$9530.21	
Postage, PHYTOPATHOLOGY	398.57	$9928.78	
Secretarial work and office expenses, Editor in Chief		352.25	
Secretarial work for Business Manager		325.00	
Secretarial work for Advertising Manager		112.87	
Stamps and envelopes		36.58	
Supplies and furniture		41.20	
Printing		25.75	
Postage, 30-Year Index (part)		8.25	
Insurance on back volumes		3.04	
Reinvestment of funds, Sinking Fund		1000.00	
Refund, subscription		6.00	
Check returned by bank		6.50	
Total expenditures			$11846.22
Balance on hand:			
Checking account		2091.41	
Northwestern Federal Savings and Loan		4658.88	6750.29
			$18596.51

The 30-Year Index. In September, 1941, an edition of 1000 copies was printed. From a running inventory of orders we have recorded shipment of 672 copies, and 14 copies are being held for future shipment to members and subscribers in occupied or enemy countries. Allowing for copies damaged or lost in transit, this should leave approximately 300 copies on hand at the printers'. A summary of receipts and expenses, included in financial reports for 1941, 1942, and 1943, follows:

Receipts:	
Sales of index, April, 1941, to November, 1943, inclusive	$3271.38
Expenses:	
Honoraria to editor and collaborators	762.50
Printing and distribution	2084.50
Other expenses	364.80
	$3211.80
Receipts in excess of expenditures	59.58

Figures given do not take into consideration accounts payable or uncollectable accounts.

Report of the Auditing Committee, as of December 1, 1943. We have examined the books and find them correct, with entries completely supported by vouchers. We wish to congratulate the Business Manager and Mrs. Meier on the clear presentation they have given of the financial transactions.

SEARS P. DOOLITTLE, *Chairman*
H. A. RODENHISER

Report of the Advertising Manager for 1943. During the year 1943 there were 108 revenue-producing advertisements carried in PHYTOPATHOLOGY. These advertisements occupied 71 pages and consisted of 43 full-page, 47 half-page, and 18 quarter-page insertions. Seventeen commercial companies used PHYTOPATHOLOGY as an advertising medium in 1943. Gross advertising contracts totalled $1265.00. Net profit to the journal for the year, after cost of printing, secretarial expenses, agency commissions and discounts are deducted, will be approximately $750. The journal would be printed regardless of advertising space contracted for in each issue, hence the profit figure given above is the difference between the cash receipts from advertising and the estimated additional cost.

In addition to the above revenue-producing insertions, 22 nonrevenue-producing advertisements were carried. These consisted of 4 full-page, 10 half-page, and 8 quarter-page insertions relative to Phytopathological Classics, the Society Clearing Agency, the 30-year Index, and Biographical Memoir of Erwin F. Smith. Costs of inserting the nonrevenue-producing advertisements were included in determining the net profit from advertising.

The management has been considerably chagrined by the lack of cooperation and consideration shown by members of the Society directed towards securing advertisements. From the above figures it can readily be seen that advertising contracts could, with member cooperation, be great enough to aid materially in offsetting the expenses of publishing the journal. A plea for cooperation was made at the Thirty-Second Annual Meeting, but during 1943, only one lead for prospective advertising was referred to the Advertising Manager. Certainly the members of the Society are capable of greater coordinated effort and the management of PHYTOPATHOLOGY earnestly asks for better support in 1944.

S. L. HOPPERSTEAD,
Advertising Manager

Report of the Editor in Chief. Volume 33 of PHYTOPATHOLOGY, exclusive of its index, contains 1218 pages of printed matter and illustrations, classified as follows: One hundred eighteen articles, 49 notes, a report of the Columbus meeting of the A. P. S. Council in February, 2 reports of the War Committee, 3 reports of the Committee on Standardization of Fungicidal Tests, 2 book reviews, 139 abstracts, 250 text figures and 1 plate.

From Dec. 10, 1942, to Nov. 30, 1943, a total of 165 manuscripts of articles, notes, book reviews, and reports were submitted for publication. Five manuscripts were either recalled by their authors or returned to them as unsuitable for publication in our Journal. There are now (Nov. 30) in press 34 articles and 53 abstracts. Other manuscripts on hand, as of that date, total 578 typewritten pages. It is estimated that there are on hand and accepted a sufficient number of manuscripts to fill about 660 printed journal pages. Of these manuscripts a considerable number, amounting to 102 pages, are in process of revision by their contributors.

The quality and importance of PHYTOPATHOLOGY as a research journal depend in very large measure upon the character and scientific excellence of the papers we contribute. That there has been an evident improvement over the years none can deny; still, there is room for improvement. Much of the delay in publication of approximately 20 per cent of all manuscripts ultimately accepted by the editors of our Journal is directly chargeable to the contributors. This becomes clear when we realize that final acceptability of such manuscripts has necessitated in some instances as many as three and four round trips between author and editor.

By increasing the number of pages per journal number we have been able to shorten the interval between date of acceptance and date of publication from 9.2 months in June, 1943, to 7.5 in November. The outlook for a further shortening is promising, partly because of the temporary policy of increasing the number of pages per number and in part because of the rather marked decline in receipt of manuscripts. We have thus far been fortunate in being able to receive our journal with remarkable regularity, and singularly free from the annoyances and embarrassments attendant upon such labor, material, and other conditions as are peculiar to such an extraordinary upheaval as the world is now experiencing.

H. B. HUMPHREY

Report of the Manager of Phytopathological Classics for the year 1943. I beg to submit herewith the annual report of my stewardship as Manager of Phytopathological Classics.

Report for the fiscal year beginning December 1, 1942, and ending December 1, 1943

Classics No. 1:	On hand 12–1–42	34	
	Sold during year	11	
	On hand 12–1–43		23
Classics No. 2:	On hand 12–1–42	234	
	Sold during year	11	
	On hand 12–1–43		223
Classics No. 3:	On hand 12–1–42	326	
	Sold during year	12	
	On hand 12–1–43		314

Classics No. 4:	On hand 12–1–42	387	
	Sold during year	13	
	On hand 12–1–43		374
Classics No. 5:	On hand 12–1–42	626	
	Sold during year	12	
	On hand 12–1–43		614
Classics No. 6:	On hand 12–1–42	712	
	Sold during year	16	
	On hand 12–1–43		696
Classics No. 7:	On hand 12–1–42	762	
	Sold during year	44	
	On hand 12–1–43		718

Receipts:

Cash balance on hand 12–1–42	$358.72	
Receipts during year	91.10	
Total		$449.82

Expenditures:

Postage	$ 7.25	
Journal	.29	
Total expenditures		$ 7.54
Balance on hand December 1, 1943		$442.28
Due on accounts 12–1–43		17.25

H. H. WHETZEL

Report for 1943 of the Representatives on the National Research Council. Energetic activity characterized the year's endeavor of the Division of Biology and Agriculture of the National Research Council to serve the Nation's war effort and to meet present and future problems growing out of the world conflict. Most of the Division's many committees were functioning with accelerated momentum on wartime objectives. A number of important conferences were called during the year to consider various problems growing out of the war. The following necessarily limited report covers only partially the Division's work.

At the Annual Meeting held April 10, 1943, in Washington, D. C., on nomination from the Society, J. C. Walker was elected to succeed Howard P. Barss as representative for three years beginning July 1, 1943. Robert F. Griggs and A. L. Maynard were reelected Chairman and Vice Chairman of the Division. R. E. Coker, A. C. Redfield, and Howard P. Barss were elected to the Executive Committee. S. E. A. McCallan of this Society was nominated to serve on the Board of Governors of the Crop Protection Institute to succeed H. T. Cook, now in overseas service with the Navy.

At this meeting major consideration was given to the crippling effect of current Selective Service procedures on technological wartime services in the fields of biology and agriculture. No clear solution of this serious problem was presented but the Division recommended the establishment of a committee to work with the proper war agencies on critical manpower shortages in these fields. J. S. Nicholas and J. G. Leach were later charged with this responsibility. It also recommended the establishment of a committee to work with appropriate war agencies in organizing effective research on problems arising in the war effort which involve the biological sciences. Attempts made later to find means to set up such a biological research committee were not successful. A new general National Research Council Committee on Scientific Personnel was, however, set up. The members are: H. A. Barton, Chairman, L. P. Eisenhart, R. F. Griggs, W. W. Rubey, and H. Marston Morse. Funds contributed by scientific organizations provide the committee with a working staff under Dean H. L. Dodge.

An interesting discussion, led by E. J. Kraus, dealt with the need for more adequate basic training as preparation for agricultural research. This grew out of a recommendation of the N.R.C. Fellowship Board that the Division set up a committee to encourage better training for research in agriculture. The committee personnel is: Richard Bradfield, Chairman, J. H. Gourley, C. B. Hutchison, J. L. Lush, and E. C. Stakman.

The Division was pleased to announce completion of projects, which included E. D. Merrill's manual of food plants and poisonous wild plants for the Pacific Islands, the manual of plants of the Central American-Caribbean region by P. C. Standley and B. E. Dahlgren, and Standley's manual of plants for the Arctic. Publication was in the hands of the War Department. The need for these manuals is evidenced by the fact that no less than 19 separate agencies of the armed forces, mostly without adequate technical help, had started efforts along such lines.

The Crop Protection Committee, through its acting chairman, J. G. Leach, reported on its joint meeting with the War Committee of the Society at Columbus, Ohio, in February. The ideas exchanged were helpful to entomologists and plant pathologists alike. A fine spirit of cooperation between them was noted. A manpower survey was reported under way by both groups. A profitable symposium, led by C. I. Bliss, on coordinated dosage experiments to determine the best way to economize on critical fungicides and insecticides was sponsored by the Committee. The Committee reported efforts toward a strengthening of Federal crop pest and disease information services. The need was presented for a study of the influence of wartime cropping changes over wide areas on the insect and disease situation.

Under the chairmanship of E. C. Stakman, the Crop Protection Committee met again in August with the Society's War Committee and Upper Mississippi Valley Group at Purdue University. The encouraging insecticide and fungicide supply was reviewed. Resolutions were presented for wartime intensification of barberry and buckthorn eradication and for all possible activity against further spread of the "Dutch" elm disease. J. S. Houser was appointed to succeed the late W. P. Flint on the Committee.

At its April meeting the Division reaffirmed its belief that "Societies in the field of science and technology represented in the Division of Biology and Agriculture of N.R.C. should plan to hold their annual meeting; that such meetings preferably not be held during Christmas holidays; that at least one session be devoted to the discussion of war problems."

D. M. Whitaker of Stanford University served the Division as Executive Secretary from February to the autumn of 1943, devoting much energy to the difficult and still unsolved problems of manpower and of mobilizing biological science for the war effort.

Throughout the year attention was given by the Division, under the chairman's leadership, to the question of how scientists in all fields of biology and agriculture can best marshall their combined strength to meet the Nation's wartime and post-war needs. Work on this problem of organization is still in progress.

The National Research Council Food and Nutrition Board, F. G. Boudreau, Chairman, continued its important activities. The Board's many committees carried out work on National wartime nutritional and food enrichment standards and on many other important scientific undertakings in the food and nutrition field.

The Committee on Animal Nutrition, under L. A. Maynard's chairmanship, with its subcommittees, was active and working closely with the Food and Nutrition Board. A Committee on Animal Health was appointed with G. H. Hart as chairman.

The Committee on Instruction in the Biological Sciences met in Cincinnati in January, 1943, and reported in favor of separate introductory courses in botany and zoology with some field work in each. E. L. Stover is Chairman.

J. C. WALKER
HOWARD P. BARSS

November 15, 1943.

Report of Committee on Donations and Legacies. During 1943 an attempt was made to interest members of the Society in investing sums of which they were capable in War Bonds and Stamps in the name of the A.P.S. Appeal letters were forwarded to all members on February 10, 1943, and May 27, 1943. The response has not been gratifying. From 23 members, bonds and stamps having a cash value of $543.00 have been received. This represents an average of approximately 50¢ per member. This is not a good record. The record on percentage of contributors is even worse; only 2 tenths of 1%. Surely, this is not indicative of the best that our profession should voluntarily produce.

The committee is gratified to have added this small amount to our endowment funds during the year, but sincerely disappointed over the response of the membership.

We recommend that a further appeal be made in the coming year, urging members of A.P.S. to support the Endowment of "Phytopathology" through the agency of War Bonds and Stamps.

E. C. STAKMAN, J. G. BROWN, N. J. GIDDINGS, NEIL STEVENS,
RICHARD P. WHITE, Chairman

Report of the Extension Work and Relations Committee. The Extension Work and Relations Committee in 1943 sponsored an open, informal discussion session at the December meeting of the Society. This session had for its objectives aiding members in plan-

ning extension projects for 1944, and also planning a program of work for the committee.

In addition, through its War Emergency Subcommittee, the following activities have been carried on:

(1) A Kodachrome transparency exchange has resulted in the preparation and distribution of duplicate sets totaling 3,818 slides on vegetable diseases and 1,197 slides on fruit diseases. It is hoped to continue this line of work on other crops.

(2) A manual of extension methods in plant pathology was prepared by a group of pathologists under the editorship of Dr. Haskell, and distributed to plant pathologists and heads of extension work throughout the United States and Canada.

(3) A manual of disease control practices has been prepared and is awaiting a means of publication.

(4) Since the last published report of this committee a survey of extension work in plant pathology throughout the United States shows that 16 of the 37 stations filling in the questionnaires have no organized extension projects in plant pathology, while 21 have. Results from this questionnaire were given at the 1942 summer meeting at Toledo.

At the summer meeting of the War Emergency Committee at Lafayette, the committee sponsored a movement to revive the EXTENSION PATHOLOGIST.

O. D. BURKE

Report of the Necrology Committee. During 1943, there were three deaths within our membership as follows:

ANDRÉS R. LÓPEZ, February 4.
ISAAC MCKINNEY LEWIS, March 12.
H. E. PARSON, November 18.

A. G. JOHNSON, Chairman
M. B. WAITE

Report of the Public Relations Committee for 1943. This committee has continued to function along lines presented in the report for last year. The efforts expended by the individual members have been greater than during the previous year with less apparent success. In some instances excellent cooperation with editors and publishers in agricultural journals and in the newspaper field has resulted in the use of material on plant pathology. Generally, however, the response has been poor, possibly because of the lack of proper preparation of the articles, possibly because this type of material has been crowded out entirely by war news and appeals for production efforts. The Committee solicits the services of members who have ability in this publicity field.

All numbers of the journal have been promptly handled, and the same responsibility can be expected for the future. Any suggestions or constructive criticisms aimed at the improvement of the projected work is requested and appreciated.

The Committee wishes to thank all who have contributed toward its functioning during the year.

L. M. BLANK, O. C. BOYD, K. S. CHESTER, J. J. CHRISTENSEN, C. T. GREGORY, J. H. JENSEN, F. P. MCWHORTER, A. G. NEWHALL, J. A. PINCKARD, L. SHAW, G. H. STARR, C. E. YARWOOD, G. F. WEBER, Chairman

Report of the Committee on Regulatory Work and Foreign Plant Diseases. The plant quarantine aspect of the outbreak of the so-called golden nematode (*Heterodera rostochiensis* Woll.) in Long Island, N. Y., in 1941 is of outstanding interest. This soil parasite seems to be spreading locally with some persistence and presents a quarantine problem which ought to be met promptly and studied with care from every angle.

As a matter of possible interest, this committee has prepared a summary of the various proposals and recommendations made by the Society in the quarantine and regulatory field during the past 15 years. A copy of this review is being filed with the Secretary and further copies can be obtained by members who may have an interest in the subject. It is suggested that some report on the progress or status of these items be presented to the Society at its next meeting. Your committee suggests that there be added to this existing list of subjects to which the Society has given and will continue to give profitable attention the following:

(1) Bacterial diseases as a foreign plant quarantine problem.

(2) The advisabiilty of making arrangements to have certain imported field and vegetable seeds treated before distribution here, as a protective measure parallel with and complementary to the domestic seed treatment measures advocated by the Society's War Board.

(3) A discussion of the value of certification in the country of origin in protecting this country against foreign disease introduction.

This Society has on several occasions expressed a deep interest in proposals for potato wart eradication from the limited areas in the three states in which it is known to occur. Intimately connected with this proposal, however, is the need for determining the correct

national policy which ought to be adopted toward the importation of foreign potatoes, either from the standpoint of potato wart alone, or in a larger way to include consideration of all foreign potato diseases and insects with a view to securing as high a degree of protection as possible for one of this country's basic food crops. In view of the probability that post-war trade resumption may involve demands for increased foreign potato importation it is important that our foreign plant quarantine policy towards potatoes be established now on a sound basis as an adequate guide to future quarantine administration. It is believed that the Society can contribute usefully to the country's welfare and render a definite service to the federal administration by giving its best thought to both aspects of the wart problem, and it is recommended that a committee be constituted either by suitably enlarging the present committee or by the appointment of a separate committee, as the Society may desire, to study the subject and report with recommendations at an early date. It may be assumed that all available information will be placed at the disposal of such a committee.

C. R. ORTON, R. P. WHITE, E. C. STAKMAN, Chairman

Report of the Committee on Biological Abstracts and the Union of American Biological Societies. BIOLOGICAL ABSTRACTS has successfully passed another milestone in its difficult wartime course. Though still a chronic issue, the financial situation now takes second place before the problem of personnel maintenance. Fortunately the editors, Dr. John E. Flynn and Dr. Jean MacCreight, as well as H. I. Anderson, business manager, continue to stand by the ship at a considerable personal sacrifice. To them the biological fraternity is deeply indebted. The resignation of several key members of the bibliographic and clerical staff, however, has had its adverse effect, particularly because of replacement difficulties. As a result, the central office staff has shrunken to four less than the number in 1940. Furthermore, the resignation of collaborators entering the armed forces, frequent changes of address, and the preoccupation of so many of the trustees, section editors and collaborators with pressing wartime duties have likewise added very appreciably to the difficulties encountered.

Nevertheless, Biological Abstracts has continued full speed ahead. The 1943 volume will contain 25,947 abstracts—2500 over 1942 in spite of evident shrinkage in many important journals. Phytopathologists will be interested in the fact that abstracts in the plant disease field number 956 in 1943—over 11 per cent more than the year before. The current increase for the complete edition is mainly accounted for by the extension in coverage to some 1800 periodicals—about 100 more than in 1942, largely because those of the Axis and German-occupied Europe are now becoming available through microfilm.

The separately published subject-matter sections continue to increase in popularity. The new Section G—Abstracts of Food and Nutrition Research is already firmly established. Subscriptions to sections have increased 24 per cent and to the complete edition 10 per cent, the total reaching an all-time high of 3634 paid subscriptions, or 19 per cent above 1942. Subscriptions to Section D—Abstracts of Plant Science have reached 455 (382 domestic, 73 foreign). By the end of October, some 13 new subscriptions had been received from members of the Society as a result of circularization in late summer. With respect to the plant disease material in Biological Abstracts, the Editor-in-Chief has this to say: "In the field of phytopathology, it is a pleasure to acknowledge that whatever success we may have had in the publication of scholarly abstracts and indexes is mainly due to the fine and careful work of its editor, Doctor Freeman Weiss. Doctor Weiss has labored with rare devotion, and the scholarship such as one sees all too infrequently. He deserves the Society's thanks for his services as editor given wholly without compensation."

Biological Abstracts of course still proceeds under considerable financial difficulty. The support of American biologists, as reflected in abstracting done and in willingness to work for the success of the whole enterprise, continues to be outstanding and their backing as expressed in financial terms is likewise excellent and on the increase. Nonetheless the income from subscriptions falls considerably short of what is required to produce Biological Abstracts. In 1942, $5,436 had to be withdrawn from the reserve. In 1943, thanks to contributions from biological societies estimated at over $3,500, and to donations from some 50 corporations already totaling nearly $9,000, draft on the reserve may perhaps be avoided. It should be emphasized, however, that grants from industrial concerns are not necessarily recurrent. It is therefore to increased subscription income that the trustees must look, in the main, for the funds that will put the enterprise on a sound sustaining basis. Not only the trustees and editors of Biological Abstracts, but the whole biological fraternity, owe gratitude to the biological societies and industries whose contributions are helping to maintain the service through this difficult period.

As they compare what has been accomplished with the needs for building up an abstracting and fact-gathering service that would more completely serve the nation's interests and the requirements of its biologists, the editors of Biological Abstracts recognize that present achievement is still far from the goals they have in mind. Every pos-

sible effort, however, is being put forth to obtain more and more of the European publications and to get them promptly and expertly abstracted, although this must now be done largely from microfilm copy. Indexing on the scale required for Biological Abstracts is a task of enormous complexity and difficulty—a job for experts in each subject matter field. Considerable progress has been made during the year in developing a staff of indexers and in working out techniques that should enable publication of future indexes at progressively earlier dates. The trustees and editors feel a great responsibility for maintaining the continuity of this important service through the present world catastrophe and beyond. In proportion as the backing is forthcoming can its performance be enlarged and improved.

FREDERICK V. RAND, Chairman

Nov. 6, 1943

Resolutions of the Committee on Resolutions. RESOLVED that The American Phytopathological Society convey to the management and staff of The Neil House, and especially to Mr. V. C. Murphy and Miss Kathryn Weitzel, its appreciation for the courteous service extended to our members and for cooperation in providing our officers and committees with unusually commodious quarters for conducting business and scientific programs.

RESOLVED that the Society express to the Committee on Local Arrangements, Dr. W. G. Stover, C. W. Ellett, R. U. Swingle and H. E. Reed, our gratitude for their fine contribution to the success of the meeting.

RESOLVED that this Society commend Dr. H. A. Edson, our Treasurer and Business Manager, for his very efficient administration of the affairs of the Society, extending through nearly a decade, and for his tireless efforts and his keen business insight reflected in our Journal, and the Society's investments, which have flourished as never before in the history of the Society.

RESOLVED that the Society commend Mrs. Agnes E. Meier, Assistant Business Manager, for her very efficient work in serving the best interests of the members of the Society.

RESOLVED that the Society express their gratitude to the Botany Department of the Ohio State University for very kindly placing at the Society's disposal projection apparatus, its good offices, laboratories and personnel. To the Horticultural Department our appreciation for the beautiful bouquet of flowers that graced our banquet table.

RESOLVED that the Society express its appreciation to Professor E. S. Thomas for his very interesting and educational illustrated lecture on "Outdoor Ohio in Natural Color."

RESOLVED that this Society on behalf of its members express to the officers and members of committees our sincere appreciation for the efforts they have made to promote the interests of the Society throughout the year, and to make this annual meeting instructive and enjoyable.

Respectfully submitted,

CHARLES CHUPP
I. E. MELHUS
C. M. TUCKER, Chairman

ACTION TAKEN BY THE COUNCIL AND SOCIETY AT THE 1943 ANNUAL MEETING, AND THROUGH MARCH 1, 1944

Elections and Appointments. The election committee opened and counted the ballots and the results were announced to the Society at the banquet. The names of those elected and appointed appear earlier in this report in the list of officers, representatives, and committees.

Thirty applicants were elected to membership in The American Phytopathological Society.

Following the 1943 annual meeting, the Editor-in-Chief of PHYTOPATHOLOGY, Dr. H. B. Humphrey, submitted his resignation to the Council which the Council accepted. The Council appointed Dr. Helen Hart as Editor-in-Chief of PHYTOPATHOLOGY to fill the unexpired term (through 1945) subject to the approval of the Society at the next annual meeting.

W. H. Burkholder, A. J. Riker and W. J. Zaumeyer were appointed editors of PHYTOPATHOLOGY, and A. A. Dunlap, Helen Hart, W. G. Hoyman and H. N. Racicot were appointed associate editors of PHYTOPATHOLOGY—all for a three year term.

R. M. Caldwell was appointed Business Manager of PHYTOPATHOLOGY for a three year term, and S. L. Hopperstead, Advertising Manager, for a one year term.

Reports of Officers, Representatives, and Committees. The Council recommended acceptance of the reports, which were approved by the Society. Reports of officers, representatives, and standing committees are published on the previous pages. According to action of the Society at the Philadelphia meeting only reports of the standing committees are to be published in the annual report.

Tropical Research Foundation. The Society confirmed the Council's recommendation to accept the final report of our representative, Dr. R. D. Rands, with appreciation of the active work carried on by our representative and that the representative be excused from further service. The announcement of the "Dissolution of the Tropical Plant Research Foundation" was published in an earlier issue of PHYTOPATHOLOGY.

Special Committee—Coordination of Cereal and Vegetable Seed Treatment Research. The Society confirmed the Council's recommendation that the names of members of the sub-committees of the Special Committee on "Coordination of cereal and vegetable seed treatment research" not be published in PHYTOPATHOLOGY. The chairman of this committee was authorized to add members to this committee when necessary during the year. The Council expressed its appreciation in the name of the Society for the faithful and important work that this committee is doing.

Publication of Monographs. The Council recommended and the Society approved that the Special Committee on "Publication of Monographs" be discharged.

Recommended Methods. A report on "recommended methods" by the committee on "Standardization of fungicidal tests" was accepted by the Society, and the Society confirmed the recommendation of the Council to refer this to the Editor-in-Chief of PHYTOPATHOLOGY for consideration relative to publishing in PHYTOPATHOLOGY.

Phytopathology. The Council recommended and the Society approved the appointment of a Special Committee to include the Business Manager of PHYTOPATHOLOGY and the Editor-in-Chief as ex officio members "to make a study of the problems affecting the publication—PHYTOPATHOLOGY."

Membership List. It was moved by the Society that a membership list be published in 1944—the matter of time and how to be left to the discretion of the officers of the Society. President J. J. Christensen appointed a Temporary Committee which has reported on the matter of publishing a membership list, and the report has been accepted by the officers of the Society.

1944 Society Meeting. The Society approved the recommendations of the Council that no official summer meeting be held, but that an annual meeting be held in 1944—the time and place to be decided by the program committee. Since this action of the Society the Council has voted not to hold the annual meeting with AAAS, September 9–16, 1944, Cleveland, Ohio, but to hold a separate meeting early in December when more field data will be available.

ANDRÉS R. LÓPEZ ELÍAS

February 4, 1909–February 4, 1943

Andrés R. López was graduated from the Polytechnic Institute at San Germán, Puerto Rico, in 1928. During 1928–29, he attended the college of Agriculture and Mechanic Arts, University of Puerto Rico, and then transferred to Louisiana University where he received the degree of Bachelor of Science in Agriculture in 1933 and the degree of Master of Science in 1935, majoring in Plant Pathology. During 1939–40, he pursued further graduate study at Iowa State University.

For a time Mr. López served as Agronomist in the Federal Emergency Relief Administration and later taught vocational agriculture in one of the Second Unit Schools of the Department of Education at San Sebastian, Puerto Rico. From 1936 to the time of his death, except for the school year 1939–40, he served as Assistant Pathologist at the Agricultural Experiment Station of the University of Puerto Rico.

Mr. López was a young man of great promise in his chosen field of science and a true friend among his colleagues.

HOWARD EVERETT PARSON

May 13, 1897–November 18, 1943

Howard Everett Parson was graduated from Michigan State College in 1923 with the degree of Bachelor of Science and received the degree of Master of Science from the University of Minnesota in 1928.

During the school year of 1924–25, Mr. Parson was superintendent of and taught agriculture in the Smith-Hughes Consolidated School at Mesick, Michigan. From 1925 to 1928, he was Instructor in Botany and Assistant in Plant Pathology in the University of Minnesota. In 1929, he was appointed Junior Pathologist in the Bureau of Plant Industry, U. S. Department of Agriculture, and worked on diseases of pecans. In 1930, he was advanced to the rank of Assistant Pathologist, and later was placed in charge of investigations of pecan diseases.

Mr. Parson was a man of quiet, retiring disposition, unusually conscientious and dependable in his work. He was unusually congenial in his associations, and made friends wherever he went.

STAUFFER SULPHURS

SULPHUR

"3-M" Magnetic Micron Measured Sulfur (Dry Wettable)

"Magnetic-Spray" Wettable Sulfur

"Magnetic" Catalytic Sulfur

"Magnetic" Humidust

"Magnetic" Cryolite

"Magnetic" Cryolite Dusts & Sprays

"Magnetic" Carbon Bisulphide

"Magnetic" Carbon Bisulphide Emulsion

"Crown" Brand Wettable Sulfur

"Electric" Super-Adhesive Dusting Sulfur

"Swan" Brand Superfine Ventilated Dusting Sulfur

"Perfection" Brand Dusting Sulfur

"Owl" Brand Superfine Dusting Sulfur

Tartox (Tartar Emetic)

"Toro" Brand Agricultural Soil Sulfur

"Triangle" Brand Commercial Flour Sulfur

Borax

STAUFFER

STAUFFER CHEMICAL COMPANY

420 Lexington Ave., New York 17, N. Y. Chicago 11, Ill.
Houston 2, Texas 624 California St., San Francisco 8, Calif.
N. Portland, Oregon Apopka, Florida

Stauffer CHEMICALS SINCE 1885

STAUFFER • SULPHUR

TRIARCH WAR ECONOMY:

There are no shortages in any materials used to make Triarch microscope slides or preserved plants, and you will not hinder the war effort by purchasing them now.

Why not build up your teaching stock of microscope slides while they are still available at pre-war prices and while most other laboratory equipment is not available?

Send for TRIARCH CATALOGS:

No. 6, with over 200 preserved plant items, 350 regular slide items and many specials.

No. 6A, special slide sets at special low prices.

No. 6B, slides of economic plants and their diseases.

GEO. H. CONANT
TRIARCH BOTANICAL PRODUCTS
Ripon, Wisconsin

"Black Leaf 40"

Aphids, Leafhoppers, and similar insects transmit many diseases including Fire Blight on apples, pears, and quinces; Black Rot and Mosaic on cabbage, cauliflower and mustard; and others equally destructive.

Spray or Dust with **BLACK LEAF 40—**

Combat these insects—

Reduce losses from both insects and diseases with one material.

Tobacco By-Products & Chemical Corp.
Incorporated
Louisville, Ky. Richmond, Va.

"Black Leaf 40"